EURO EVO DEVO

22–25 July 2014 l Vienna, Austria

Previous Meetings

2006 Prague

2008 Ghent

2010 Paris

2012 Lisbon

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Program Committee

Frietson Galis (Chair) – VU University Medical Centre, Amsterdam & NCB Naturalis
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Ronald Jenner – Natural History Museum, London

Welcome from the president

"Servus" to all participants of the 5th meeting of the European Society for Evolutionary Developmental Biology!

Since its founding in 2006, our society has grown steadily and so did the number of attendees at its biennial conferences. This time, nearly 600 evolutionary developmental biologists from all over the world will enjoy three days of exchange and open discussion of the latest research in the field. EvoDevo has matured into a robust scientific discipline, analyzing developmental evolution at all levels of scale, from molecules and proteins to complex structures and populations. It is this broad interpretation of EvoDevo that our society intends to foster. Therefore, this year, in addition to many experimental subjects, the topics include theory, history, and philosophy of EvoDevo, as well as quantitative, bioinformatic, and behavioral EvoDevo. It will be exciting to hear from all these different perspectives.

The extraordinary number of registrations for the meeting indicates a healthy proliferation of EvoDevo research, but it also has its downsides. More parallel sessions had to be established, and the posters will have to be displayed in two separate locations. At certain times, movement between lecture halls may become congested. We ask you to take this as an indication of high interest in our field of science, and we beg your pardon for any inconveniences that may arise. The number of events associated with the EED conference has also increased. In addition to the traditional Tribolium satellite meeting, this time there will also be an Amphioxus satellite meeting. Both are held a day prior to our conference.

We are grateful to the sponsors who have provided support for various activities and to the conference management team of the University of Vienna for their very professional attitude. We would also like to thank the members of all committees for working hard to make this event possible. In particular, we are most grateful to Isabella Sarto-Jackson from the KLI Institute and the local organizing team for their enthusiasm and the many hours they have invested in the preparation of this meeting.

We wish you all an inspiring and exciting conference – enjoy Vienna!

On behalf of the Executive Committee,

Gerd B. Müller
President EED

General Information

Dates & Venue

Tuesday 22 to Friday 25 July 2014
Campus of the University of Vienna
Spitalgasse 2 (Hof 2)
1090 Vienna, Austria

Registration

The registration office is in the Aula (see map). If you have registered previously, it will be possible to pay the registration fee on-site. Social events during and after the conference can also be booked and paid on-site.

Opening hours

Monday 21 July	12.00 – 16.00
Tuesday 22 July	14.00 – 19.00
Wednesday 23 July	8.00 – 18.00
Thursday 24 July	8.30 – 18.00
Friday 25 July	8.30 – 19.00

Fees

Late registration fee	EUR 300,–
Late registration fee for students *(Student certificate required)*	EUR 250,–
Conference Dinner, 25 July	EUR 45,–
Boattrip to the Wachau region, 26 July	EUR 95,–
Art performance "Theory of Flight", 26 July	EUR 25,–

Fees include:

- Access to all sessions
- Welcome Reception at the venue
- Reception at Vienna City Hall (advanced registration was required)
- Delegates' documents (printed program booklet, pdf of abstracts on USB stick)
- Certificate of attendance
- Lunches in one of three restaurants on Campus
- Coffee breaks

Badges and security

Please wear your name badge at all times while at the congress venue and during the social events, as well as during the breaks. It is the official entrance pass to the scientific sessions, the welcome reception, and the campus restaurants for the free lunches on 23, 24, and 25 July.

Cancellation policy

Those who made cancellations more than one month prior to the start of the congress will be refunded 50% of the registration fee. Later cancellations cannot be considered.

Lunches

Please wear your badge!

Lunch is provided in three restaurants on Campus (see map). Due to the large number of participants, the restaurants may be crowded. Please move to another restaurant in case your first choice has no more seats available and take slightly longer waiting times into consideration. Lunches include 0.5 l of water, 0.25 l non-alcoholic drinks, a soup, and one of two main courses per day:

Wednesday 23 July
Schnitzel & potatoes *or* cabbage-and-noodle casserole & salad

Thursday 24 July
Rissole & salad *or* spinach strudel & herbed sour cream

Friday 25 July
Grilled perch & potatoes *or* pumpkin lasagna & salad

Social Events

Welcome Reception

The welcome reception will be held on Tuesday 22 July, at 19:00, in front of the lecture halls C1 & C2. The reception is free, but you will need to wear your badge to have access. The reception is sponsored by Springer.

Reception at Vienna City Hall

A reception sponsored by the City of Vienna will be held at Vienna City Hall on 23 July, 20.30 (see map). City Hall is within walking distance from the venue (about 10 to 15 minutes). Participation is free, but due to space limitations you need to have booked your attendance in advance. If you have registered for the reception, you will obtain an official invitation at the reception desk. Please bring your invitation as well as your conference badge to ensure access.

Conference Dinner

The conference dinner will take place on 25 July, 19:30, at the traditional Viennese "Heurigen" Schübel-Auer (for directions refer to the map at the end of the booklet). Special "Euro Evo Devo" trams will take registered participants to the Heurigen. Two trams will wait at the tram stop at the corner Alser Strasse/Spitalgasse at 19.30.

Boattrip to the Wachau

There are still vacancies for the boat trip on the Danube on 26 July. The destination is the Wachau region, a UNESCO World Inheritance site. If you would like to participate, please consult the registration desk.

"Theory of Flight" *art performance at the KLI Institute in Klosterneuburg*

Boston artist Anna Lindemann will perform her piece "Theory of Flight" on 26 July at 17:00. Anna Lindemann is devoted to integrating art and science. Her work combines animation, music, video, and performance to explore the emerging field of EvoDevo. She received an MFA in Integrated Electronic Arts from Rensselaer Polytechnic Institute and a BS in Biology from Yale University. Please register early if you intend to participate. The announcement in your delegate's bag has a map showing how to reach the KLI Institute.

Technical Information

WiFi Access

All registered participants receive individual access to the Wi-Fi network of the University of Vienna for the duration of the conference. A password is included with the registration materials. Eduroam is also available.

Oral Presentations

Speakers are asked to transfer their presentation (Powerpoint or pdf) from a USB stick onto the PCs in the lecture rooms before their session starts: between 8.30 and 8.55 for the morning sessions, or during the breaks immediately preceding their session. Speakers may use their own laptop computer if they prefer. In this case, they should also come to the lecture room before the start of their session, as mentioned above, in order to check whether the projection is working. It is best to have a copy of your presentation on a USB stick, in case of a problem. We kindly ask chairs to be present at least 15 minutes prior to the start of their session.

The maximum duration of symposia talks is 25 minutes (including 5 minutes for questions) and 15 minutes for contributed talks (including 3 minutes for questions)

Note: Speakers in the contributed sessions C11 to C14 exceptionally have 25 minutes available (20 + 5). Due to the large number of talks and the running of up to 5 sessions in parallel, we ask speakers to keep closely to the time schedule and session chairs to be very strict in not allowing speakers to go over the allotted times.

Poster Presentations

The poster boards are suitable for posters of the A0 format (1.2 m height x 0.9 m width). All posters can remain posted throughout the meeting. Presenters should put up their posters on Tuesday evening or Wednesday morning. Please be present at your poster during the poster sessions on Wednesday (even numbers) and Thursday (odd numbers). Materials for the fixing of the posters will be available. Posters should be removed before the end of the meeting, i.e., at the latest during the coffee break on Friday afternoon.

Poster Prize Competition

Doctoral or Master's students presenting a first-author poster will take part in the poster competition if they indicated their participation upon submission of their abstract. There will be **THREE equal-first prizes** (Apple iPad-mini tablet computer) awarded by the poster competition committee. We are grateful to both BioEssays and Annals of Botany for sponsoring this competition!

Please have one or two key images from your poster ready on a USB stick for projection at the prize-giving ceremony (around 17:20 on Friday 25 July) in case your poster is one of the winners.

Live Transmission from room C1 to room C2

Due to the large audience to be expected for the Welcome Ceremony, the Keynote Lectures, the award of the Kowalevsky Medal, the student poster Prize Ceremony, and the Conference Closing there will be a live transmission from lecture hall C1 to lecture hall C2. Please move to lecture hall C2 when you notice that lecture hall C1 is full.

Program at a Glance

Preconference Meetings

Monday, July 21st

	ROOM C1		ROOM C2
13.45 – 19.00	Satellite Meeting **Amphioxus**	14.25 – 19.30	Satellite Meeting **Tribolium**

Tuesday, July 22nd

	ROOM C1		ROOM C2
09.00 – 12.00	Satellite Meeting **Amphioxus**	08.50 – 12.00	Satellite Meeting **Tribolium**
14.00 – 17.00	Satellite Meeting **Amphioxus**	13.30 – 17.00	Satellite Meeting **Tribolium**

EURO EVO DEVO 2014

Tuesday, July 22nd

	ROOM C1	ROOM C2
18.00 – 18.20	EURO EVO DEVO 2014 **Opening**	Live transmission from C1 to C2
18.20 – 19.00	Keynote Lecture (K1) **Jean-Jacques Hublin**	Live transmission from C1 to C2
19.00	**Welcome Reception at the Venue**	

Wednesday, July 23rd

	ROOM A	ROOM B	ROOM C1	ROOM C2	ROOM D
09.00 – 10.40	Symposium S1 **Mechanical development I**	Symposium S2 **EvoDevo of colour**	Symposium S3 **Phenotypic change & NGS**	Symposium S4 **Extended Synthesis**	
10.40 – 11.10	Coffee break				
11.10 – 12.25	Contributed C1 **Living fossils**	Contributed C2 **Quantitative variation**	Contributed C3 **Phenotypic change & NGS I**	Contributed C4 **Ext. Synthesis & Quo vadis**	Contributed C5 **Developmental evolution I**
12.25 – 14.00	Lunch break				
14.00 – 15.40	Symposium S5 **Mechanical development II**	Symposium S6 **EvoDevo of symmetry**	Symposium S7 **Wnt signaling**	Symposium S8 **Quo vadis EvoDevo?**	
15.40 – 16.10	Coffee break				
16.10 – 17.10	Contributed C6 **Plant EvoDevo**	Contributed C7 **EvoDevo of symmetry**	Contributed C8 **Wnt signaling I**	Contributed C9 **Mechanical development**	Contributed C10 **Complexity in gene networks**
17.10 – 17.20	Break				
17.20 – 18.00			Keynote Lecture (K2) **Veronica Grieneisen** (live transmission from C1 to C2)		
18.00 – 20.00	**Poster Session 1** (even numbers) Corridors in front of rooms C1 & C2 and room E				
20.30	**Reception at Vienna City Hall** (see map)				

Program at a Glance

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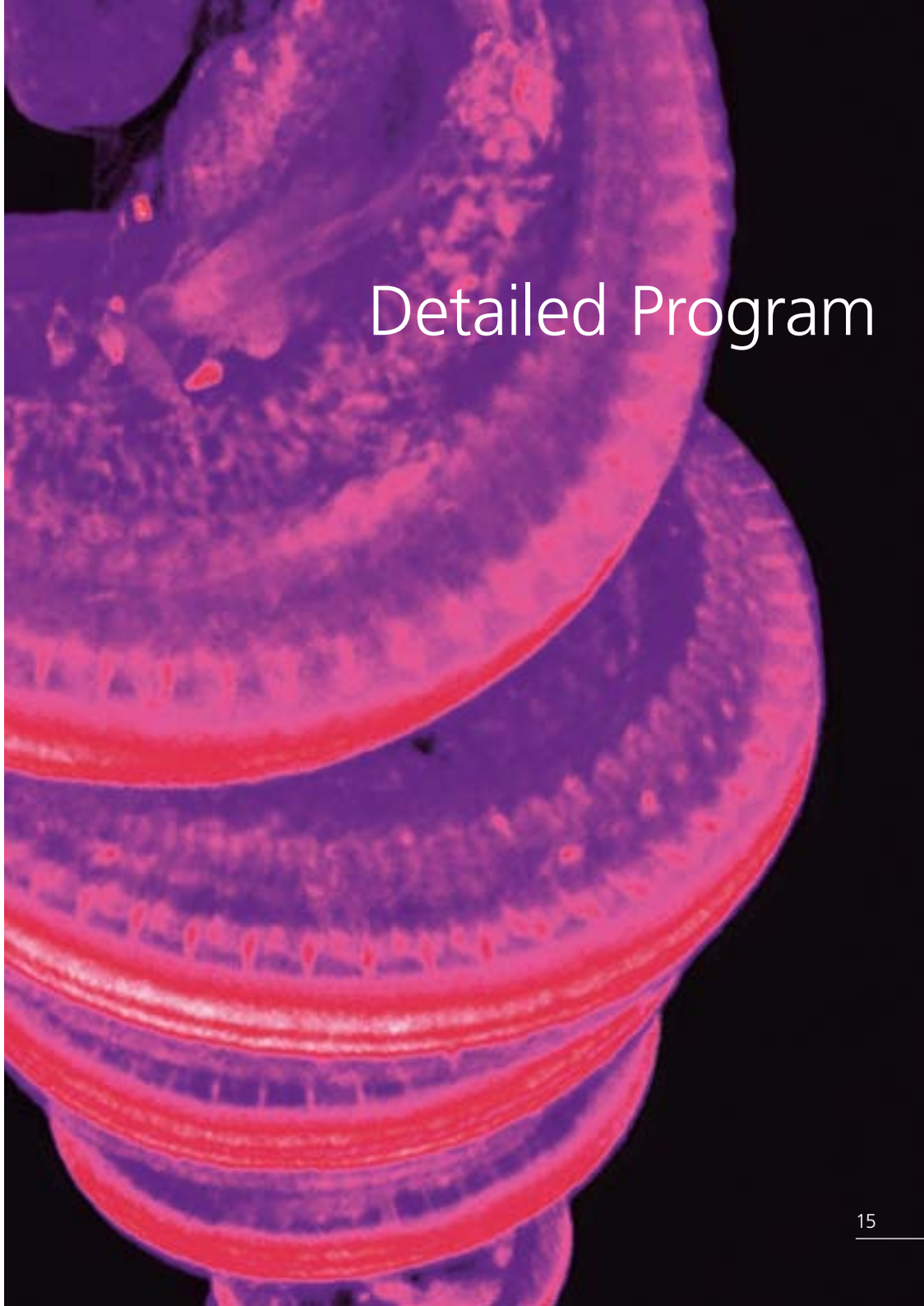
Thursday, July 24th

	ROOM A	ROOM B	ROOM C1	ROOM C2	ROOM D
09.00 – 10.40	Symposium S9 **Plant EvoDevo**	Symposium S10 **Bioinformatics in EvoDevo I**	Symposium S11 **Ecology & environment I**	Symposium S12 **Quantitative variation**	
10.40 – 11.10	Coffee break				
11.10 – 12.50	Symposium S13 **Vertebrate dentitions**	Symposium S14 **Bioinformatics in EvoDevo II**	Symposium S15 **Ecology & environment I**	Symposium S16 **Developmental robustness**	
12.50 – 14.20	Lunch break				
14.20 – 16.00	Symposium S17 **EvoDevo of communication**	Contributed C11 **Bioinformatics in EvoDevo**	Contributed C12 **Wnt signaling II**	Contributed C13 **Ecology & environment I**	Contributed C14 **Developmental evolution II**
16.00 – 16.30	Coffee break				
16.30 – 17.00	Contributed C15 **EvoDevo of communication**	Contributed C16 **Quo vadis EvoDevo?**	Contributed C17 **Regeneration EvoDevo**	Contributed C18 **Ecology & environment II**	Contributed C19 **Amphioxus EvoDevo**
17.00 – 17.10	Break				
17.10 – 17.20			Award of the Kowalevsky Medal to **Denis Duboule** (live transmission from C1 to C2)		
17.20 – 18.00			Keynote Lecture (K3) **Ulrich Technau** (live transmission from C1 to C2)		
18.00 – 20.00	Workshop **Live Imaging** (Demonstration in room F)	**Poster Session 2** (odd numbers) Corridors in front of rooms C1 & C2 and room E			

Friday, July 25th

	ROOM A	ROOM B	ROOM C1	ROOM C2	ROOM D
09.00 – 10.40	Symposium S18 **Cranial neural crest**	Symposium S19 **Quantitative EvoDevo I**	Symposium S20 **Loss of gene function**	Symposium S21 **EcoEvoDevo**	
10.40 – 11.10	Coffee break				
11.10 – 12.25	Contributed C20 **Neural crest & dentition**	Contributed C21 **Quantitative EvoDevo I**	Contributed C22 **Loss of gene function**	Contributed C23 **EcoEvoDevo**	Contributed C24 **Origin of regeneration**
12.25 – 14.00	Lunch break				
14.00 – 15.40	Symposium S22 **Marine sensory systems**	Symposium S23 **Quantitative EvoDevo II**	Symposium S24 **Origin of regeneration**	Symposium S25 **Living fossils**	
15.40 – 16.10	Coffee break				
16.10 – 17.10	Contributed C25 **Marine sensory systems**	Contributed C26 **Quantitative EvoDevo II**	Contributed C27 **Developmental robustness**	Contributed C28 **Phenotypic change & NGS**	Contributed C29 **Developmental evolution III**
17.10 – 17.20	Break				
17.20 – 17.35			**Student Poster Prizes** (live transmission from C1 to C2)		
17.35 – 18.15			Keynote Lecture (K4) **Stuart Newman** (live transmission from C1 to C2)		
18.15 – 18.20			**Conference Closing** (live transmission from C1 to C2)		
18.20 – 19.10			**EED Business Meeting**		
19.30	Joint departure for **Conference Dinner** (see map)				

Notes



Detailed Program

Detailed Program

Tuesday, July 22nd

14.00 – 18.00 Registration

18.00 – 18.20 Opening
ROOMS C1&2 **Welcome address by Gerd B. Müller**
(President of the EED and Chair of the Local Organizing Committee)

18.20 – 19.00 Keynote Lecture (K1)
Becoming Fully Human
ROOMS C1&2 **Jean-Jacques Hublin**
(Max Planck Institute for Evolutionary Anthropology, Leipzig, GER)
Chair: Gerd B. Müller

19.00 – 21.00 Welcome Reception at the Venue
sponsored by Springer

Wednesday, July 23rd

09.00 – 10.40 Symposium S1:
Mechanical mechanisms of development I
ROOM A
Organizers: Naomi Nakayama and Annemiek Cornelissen
Chairs: Naomi Nakayama and Annemiek Cornelissen

S1-01 **Patterning under pressure**
Bennett, Malcolm (University of Nottingham, GBR); Goh, Tatsuaki (Kobe University, JPN / University of Nottingham, GBR); Guyomarc'H, Soazig (IRD, UMR DIADE (IRD/UM2), FRA); Fukaki, Hidehiro (Kobe University, JPN); Laplaze, Laurent (IRD, UMR DIADE (IRD/UM2), FRA)

S1-02 **Getting the mechanics right:**
The coordination of the lateral expansion of plant stems
Werner, Stephanie (Gregor Mendel Institute, Vienna, AUT); Suer, Stephanie (Gregor Mendel Institute, Vienna, AUT); Wolf, Sebastian (Ruprecht Karl University of Heidelberg, GER); **Greb, Thomas** (Gregor Mendel Institute, Vienna, AUT)

S1-03 **Cell and tissue mechanics in zebrafish gastrulation**
Heisenberg, Carl-Philipp (IST Austria, Klosterneuburg, AUT)

S1-04 **Sequential rings of cells turn into sequential body parts and sequential evolution by physical pattern formation**
Fleury, Vincent (Laboratoire MSC/UMR7057, Paris, FRA)

09.00 – 10.40 Symposium S2:
EvoDevo of colour
ROOM B
Organizer: Beverley Glover
Chair: Beverley Glover

S2-01 **Under the rainbow: Understanding how plants build microscopic structures to produce iridescence**
Moyroud, Edwige (University of Cambridge, GBR); Vignolini, Silvia (University of Cambridge, GBR); Rudall, Paula (Royal Botanic Gardens Kew, London, GBR); Steiner, Ullrich (University of Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

S2-02 **Plant genes that alter pollinator preference**
Sheehan, Hester (University of Berne, CHE); Klahre, Ulrich (University of Berne, CHE); Dell'Olivo, Alexandre (University of Berne, CHE); Moser, Michel (University of Berne, CHE); Hermann, Katrin (University of Berne, CHE); Esfeld, Korinna (University of Berne, CHE); Summers, Holly (University of Berne, CHE); Caze, Ana Luiza (Universidade Federal do Rio Grande do Sul, Porto Alegre, BRA); Kuhlemeier, Cris (University of Berne, CHE)

S2-03 **Evolution of a novel pigmentation pattern through regulatory rewiring**
Martins, Talline (Duke University, Durham, NC, USA); Rausher, Mark (Duke University, Durham, NC, USA)

S2-04 **How chameleons change colour**
Milinkovitch, Michel (University of Geneva, CHE); Teyssier, Jeremie (University of Geneva, CHE); Saenko, Suzanne (University of Geneva, CHE); van der Marel, Dirk (University of Geneva, CHE)

09.00 – 10.40 Symposium S3: Uncovering the genomic bases of phenotypic change in the NGS era

ROOM C1

Organizers: Manuel Irimia, Ignacio Maeso, Juan Pascual-Anaya
Chairs: Manuel Irimia and Ignacio Maeso

S3-01 **NG sequencing technology and comparative genomics of non-model systems**
Hejnol, Andreas (University of Bergen, NOR); Ryan, Joe F. (University of Bergen, NOR); Vellutini, Bruno C. (University of Bergen, NOR); Martín-Durán, José María (University of Bergen, NOR); Pang, Kevin (University of Bergen, NOR); Børve, Aina (University of Bergen, NOR)

S3-02 **Mechanisms of gene regulatory evolution during the origin of mammalian pregnancy**
Lynch, Vincent J. (The University of Chicago , IL, USA)

S3-03 **Understanding extreme non-coding conservation**
Lenhard, Boris (Imperial College London, GBR)

S3-04 **Big mice on small islands: The origin and evolution of the Faroese house mouse**
Chan, Frank (Friedrich Miescher Laboratory of the Max Planck Society, Tübingen, GER)

09.00 – 10.40 Symposium S4: Extended Evolutionary Synthesis

ROOM C2

Organizers: Gerd B. Müller and Werner Callebaut
Chair: Gerd B. Müller

S4-01 **Niche construction, developmental bias and the Extended Evolutionary Synthesis**
Laland, Kevin (University of St Andrews, GBR); Uller, Tobias (University of Oxford, GBR)

S4-02 **An evolutionary view of brain development and dynamics and its theoretical consequences**
Szathmáry, Eörs (Parmenides Foundation, Pullach, GER)

S4-03 **A new vision of heredity to build an inclusive Evolutionary Synthesis**
Danchin, Etienne (CNRS, Toulouse, FRA)

S4-04 **The role of EvoDevo in extending the Evolutionary Synthesis**
Callebaut, Werner (The KLI Institute, Klosterneuburg, AUT); Müller, Gerd B. (University of Vienna, AUT)

11.10 – 12.25 Contributed Session C1: "Living fossils", myth or reality?

ROOM A

Chairs: Didier Casane and Patrick Laurenti

C1-01 **Sharks, stems, and shedding the living fossil tag**
Coates, Michael (The University of Chicago, IL, USA); Criswell, Katharine (The University of Chicago, IL, USA)

C1-02 **Skeletogenesis in cartilaginous fish**
Enault, Sebastien (ISEM - Université Montpellier 2, FRA); Da Silva, Willian (ISEM - Université Montpellier 2, FRA); Venteo, Stephanie (INM- Université Montpellier 2, FRA); Debiais-Thibaud, Mélanie (ISEM Université Montpellier 2, FRA)

C1-03 ***Limulus polyphemus* is not quite a "living fossil", especially from a developmental point of view**
Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

C1-04 **Cockroach-like insects: Successful since 300 million years and therefore "living fossils"?**
Hörnig, Marie K. (Ernst-Moritz-Arndt-University of Greifswald, GER); Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Joachim T. (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

C1-05 **Segmentation in brachiopod larvae? The expression of common segment patterning genes during development of larval lobes**
Vellutini, Bruno (University of Bergen, NOR); Hejnol, Andreas (University of Bergen, NOR)

11.10 – 12.25 Contributed Session C2:
Developmental basis of quantitative variation

ROOM B *Chair:* Mihaela Pavlicev

C2-01 **How do you shave a baby? *Cis*-regulatory region occupancy as the basis for morphological evolution**
Preger-Ben Noon, Ella (Janelia Farm Research Campus, Ashburn, VA, USA); Stern, David (Janelia Farm Research Campus, Ashburn, VA, USA)

C2-02 **Tissue sensitivity to Hox protein levels underlies the adaptive leg morphology of water striders**
Refki, Peter (Institute of Functional Genomics (IGFL); UCBL, Lyon, FRA); Armisen, David (IGFL-ENS, Lyon, FRA); Crumière, Antonin (IGFL-ENS, Lyon, FRA); Viala, Séverine (IGFL-ENS, Lyon, FRA); Khila, Abderrahman (IGFL-ENS, Lyon, FRA)

C2-03 **Developmental mechanisms underlying natural variation in organ size**
Ramaekers, Ariane (VIB-KU Leuven, BEL); Weinberger, Simon (VIB-KULeuven, BEL); Buchner, Erich (University of Würzburg, GER); Wolf, Reinhard (University of Würzburg, GER); Hassan, Bassem A. (VIB-KU Leuven, BEL)

C2-04 **Investigating the genetic and developmental origins of limb bone length using mice selectively bred for increased tibia length**
Marchini, Marta (University of Calgary, AB, CAN); Krueger, Carsten B. (University of Calgary, AB, CAN); Sparrow, Leah M. (University of Calgary, AB, CAN); Dowhanik, Alexandra S. (University of Calgary, AB, CAN); Cosman, Miranda N. (University of Calgary, AB, CAN); Rolian, Campbell (University of Calgary, AB, CAN)

C2-05 **Pattern modulation produces a highly regular grid of defensive hairs in the spiny mouse (*Acomys dimidiatus*)**
Tzika, Athanasia (University of Geneva, CHE); Montandon, Sophie (University of Geneva, CHE); Manukyan, Liana (University of Geneva, CHE); Milinkovitch, Michel (University of Geneva, CHE)

11.10 – 12.25 Contributed Session C3:
Uncovering the genomic bases of phenotypic change in the NGS era I

ROOM C1 *Chairs:* Manuel Irimia and Juan Pascual

C3-01 **Two novel, complementary next generation sequencing approaches to reveal the dorso-ventral gene regulatory network of *Tribolium castaneum***
Stappert, Dominik (University of Cologne, GER); Frey, Nadine (University of Cologne, GER); Roth, Siegfried (University of Cologne, GER)

C3-02 **Convergent evolution of proteins with repetitive, low complexity domains in biomineralizing taxa**
McDougall, Carmel (University of Queensland, Brisbane, AUS); Woodcroft, Ben (University of Queensland, Brisbane, AUS); Degnan, Bernard (University of Queensland, Brisbane, AUS)

C3-03 **Cnidarian microRNAs frequently regulate their targets by cleavage**
Praher, Daniela (University of Vienna, AUT); Moran, Yehu (The Hebrew University of Jerusalem, ISR); Fredman, David (University of Vienna, AUT); Li, Xin (University of Massachusetts, Worcester, MA, USA); Wee, Liang-Meng (University of Massachusetts, Worcester, MA, USA); Rentzsch, Fabian (University of Bergen, NOR); Zamore, Philip (University of Massachusetts, Worcester, MA, USA); Seitz, Hervé (CNRS, Montpellier, FRA); Technau, Ulrich (University of Vienna, AUT)

C3-04 **Trancriptome profiling of a key morphological innovation: The propelling fan of the water walking bug *Rhagovelia obesa***
Santos, Emilia (Institute of Functional Genomics (IGFL), Lyon, FRA); Khila, Abderrahman (IGFL, Lyon, FRA)

C3-05 **Phenotypic plasticity and epigenetics in the honeybee ovary**
Leask, Megan (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL); Duncan, Elizabeth (University of Otago, Dunedin, NZL)

11.10 – 12.25 Contributed Session C4:
Extended Evolutionary Synthesis & Quo vadis EvoDevo?

ROOM C2 *Chair:* Werner Callebaut

C4-01 **The origination of novelty: Qualitative changes from quantitative variation**
Peterson, Tim (University of Vienna, AUT); Müller, Gerd B. (University of Vienna, AUT)

C4-02 **Adaptive dynamics modelling with evolving epigenetic switches**
Van Dooren, Tom J. M. (Institute of Ecology and Environmental Sciences Paris, FRA)

C4-03 **The timing of development**
Nicoglou, Antonine (Institut d'Histoire et de Philosophie des Sciences et des Techniques, Paris, FRA)

C4-04 **How a better understand of developmental mechanisms would transform the bases of evolutionary biology**
Salazar-Ciudad, Isaac (Institute of Biotechnology, Helsinki, FIN)

C4-05 **Computing the concept of evolvability**
Nuño de la Rosa, Laura (The KLI Institute, Klosterneuburg, AUT)

11.10 – 12.25 Contributed Session C5:
Developmental mechanisms underlying evolutionary change I

ROOM D

Chair: Constanze Bickelmann

C5-01 **Hox expression in salamanders: Preaxial polarity revisited**
Bickelmann, Constanze (Museum für Naturkunde, Berlin, GER); Schneider, Igor (Instituto de Ciencias Biologicas, Belem, BRA); Fröbisch, Nadia B. (Museum für Naturkunde, Berlin, GER)

C5-02 **An embryological perspective on lung structure of early amniotes**
Lambertz, Markus (University of Bonn, GER); Grommes, Kristina (University ofBonn, GER); Kohlsdorf, Tiana (Universidade de São Paulo, Ribeirão Preto, BRA); Perry, Steven F. (University of Bonn, GER)

C5-03 **The intricate relationship between selection and developmental constraints: the evolution of the Drosophila sex comb length**
Malagon, Juan (University of Toronto, CAN)

C5-04 **A burst of microRNA innovation in the early evolution of butterflies and moths**
Quah, Shan (University of Oxford, GBR); Holland, Peter (University of Oxford, GBR)

C5-05 **Stem cell genes characterization in *Oscarella lobularis* (Porifera, Homoscleromorpha): The stepping stone towards understanding somatic and germ lines origin**
Fierro, Laura (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); Schenkelaars, Quentin (IMBE, Marseille, FRA); Borchiellini, Carole (IMBE, Marseille, FRA); Ereskovsky, Alexander (IMBE, Marseille, FRA); Renard, Emmanuelle (IMBE, Marseille, FRA)

14.00 – 15.40 Symposium S5:
Mechanical mechanisms of development II

ROOM A

Organizers: Naomi Nakayama and Annemiek Cornelissen
Chairs: Naomi Nakayama and Annemiek Cornelissen

S5-01 **Directional mechanical signals add robustness to plant morphogenesis**
Hamant, Olivier (ENS Lyon, FRA)

S5-02 **Mechanics of cell contacts during tissue morphogenesis**
Lenne, Pierre-François (IBDM, Marseille, FRA)

S5-03 **Mechanical development of veins controls shape, position and movements of plants leaves**
Douady, Stéphane (Université Paris-Diderot / CNRS, FRA)

S5-04 **Mechanical basis of seashell morphogenesis**
Moulton, Derek (University of Oxford, GBR); Goriely, Alain (University of Oxford, GBR); Chirat, Régis (Université Lyon 1, Villeurbanne, FRA)

14.00 – 15.40 Symposium S6:
EvoDevo of symmetry in animals and plants

ROOM B

Organizers: Sophie Nadot and Catherine Damerval
Chair: Catherine Damerval

S6-01 **Do asymmetric flies fly in circles? Functional consequences of a genetically induced asymmetry on flight performance in *Drosophila melanogaster***
Debat, Vincent (UMR7502 ISyEB, Museum National d'Histoire Naturelle, Paris, FRA); Aponte, Jose David (Florida State University, Tallahassee, FL, USA); Cornette, Raphaël (UMR7502 ISyEB, Museum National d'Histoire Naturelle, Paris, FRA); Herrel, Anthony (UMR7504 FUNEVOL, Museum National d'Histoire Naturelle, Paris, FRA); Peronnet, Frédérique (UMR Biologie du Développement, UPMC, Paris, FRA)

S6-02 **Role of myosin ID in Drosophila and zebrafish left-right asymmetry**
Noselli, Stephane (IBV - CNRS UMR7277, Nice, FRA)

S6-03 **Corolla monosymmetry: Evolution of a morphological novelty in the angiosperms**
Zachgo, Sabine (Osnabrück University, GER)

S6-04 ***CYCLOIDEA*-like genes and floral symmetry in Ranunculaceae**
Jabbour, Florian (National Museum of Natural History, Paris, FRA); Cossard, Guillaume (University of Lausanne, CHE); Le Guilloux, Martine (CNRS, Gif-sur-Yvette, FRA); Sannier, Julie (Université Paris-Sud, Orsay, FRA); Nadot, Sophie (Université Paris-Sud, Orsay, FRA); Damerval, Catherine (CNRS, Gif-sur-Yvette, FRA)

14.00 – 15.40 Symposium S7:
The Roche Discovery Oncology Symposium: Perspectives on Wnt signaling

ROOM C1

Organizers: Wim Damen and Cornelius Eibner
Chairs: Wim Damen and Cornelius Eibner

S7-01 **Cancer mutations derail Wnt signalling via conformational conversion of the scaffold protein Axin**
Maurice, Madelon (Utrecht University, NLD); Anvarian, Zeinab (Utrecht University, NLD); Nojima, Hisashi (MRC National Institute for Medical Research, Mill Hill, London, GBR); Madl, Tobias (Utrecht University, NLD); Spit, Maureen (Utrecht University, NLD); van Kappel, Eline (Utrecht University, NLD); Scherpenzeel, Revina (Utrecht University, NLD); Low, Teck Y. (Utrecht University, NLD); Kuper, Ineke (Utrecht University, NLD); Jordens, Ingrid (Utrecht University, NLD); Gerlach, Jan P (Utrecht University, NLD); Heck, Albert J. R. (Utrecht University, NLD); Vincent, Jean-Paul (MRC National Institute for Medical Research, Mill Hill, London, GBR); Rüdiger, Stefan G. D. (Utrecht University, NLD)

S7-02 **Wnt signaling in the annelid *Platynereis dumerilii***
Demilly, Adrien (Institut Jacques Monod CNRS, Paris, FRA); Gazave, Eve (Institut Jacques Monod CNRS, Paris, FRA); Steinmetz, Patrick (University of Vienna, AUT); Marchand, Lauriane (Institut Jacques Monod CNRS, Paris, FRA); Kerner, Pierre (Institut Jacques Monod CNRS, Paris, FRA); Vervoort, Michel (Institut Jacques Monod CNRS, Paris, FRA)

S7-03 **Wnt signaling shapes planarians**
Adell, Teresa (University of Barcelona, ESP); Sureda, Miquel (University of Barcelona, ESP); Almuedo-Castillo, Maria (University of Barcelona, ESP); Martín-Durán, José María (University of Barcelona, ESP); Rojo-Laguna, Jose Ignacio (University of Barcelona, ESP); Saló, Emili (University of Barcelona, ESP)

S7-04 **Dissecting Wnt-signaling dependent transcription in the mouse**
Cantù, Claudio (University of Zurich, CHE)

14.00 – 15.40 Symposium S8: Quo vadis EvoDevo?

ROOM C2

Organizers: Manfred Laubichler and Cassandra Extavour
Chair: Manfred Laubichler
Discussion leader: Ronald Jenner

S8-01 **EvoDevo in the Americas: A report on a community workshop to consolidate and advance the field of Evolutionary Developmental Biology**
Extavour, Cassandra (Harvard University, Cambridge, MA, USA)

S8-02 **The future of evo-devo**
Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

S8-03 **Quo vadis EvoDevo?**
Laubichler, Manfred (Arizona State University, Tempe, USA)

16.10 – 17.10 Contributed Session C6: Plant EvoDevo: Linking cross-species genetic and morphological variation

ROOM A

Chair: Caspar Chater

C6-01 **Deep homology in the land plant stomatal development programmed**
Chater, Caspar (University of Sheffield, GBR); Caine, Robert (University of Sheffield, GBR); Kamisugi, Yasuko (University of Leeds, GBR); Cuming, Andrew (University of Leeds, GBR); Fleming, Andrew (University of Sheffield, GBR); Beerling, David (University of Sheffield, GBR); Gray, Julie (University of Sheffield, GBR)

C6-02 **Through the periscope: Understanding early grass leaf development**
Richardson, Annis (John Innes Centre, Norwich, GBR); Rebocho, Xana (John Innes Centre, Norwich, GBR); O'Connor, Devin (Cambridge University, GBR); Hake, Sarah (University of California Berkeley, CA, USA); Coen, Enrico (John Innes Centre, Norwich, GBR)

C6-03 **Three ancient hormonal cues co-ordinate shoot branching in a moss**
Coudert, Yoan (University of Cambridge, GBR); Palubicki, Wojtek (Cambridge University, GBR); Leyser, Ottoline (Cambridge University, GBR); Harrison, Jill (University of Cambridge, GBR)

C6-04 **Stay high or get low: Can epigenetic variation lead to recurrent speciation?**
Paun, Ovidiu (University of Vienna, AUT); Flatscher, Ruth (University of Vienna, AUT); Frajman, Bozo (University of Innsbruck, AUT); Trucchi, Emiliano (University of Vienna, AUT); Schönswetter, Peter (University of Innsbruck, AUT)

16.10 – 17.10 Contributed Session C7: EvoDevo of symmetry in animals and plants

ROOM B

Chair: Sophie Nadot

C7-01 **A computational model of cleavage patterns in metazoa and its evolution**
Brun-Usan, Miguel (Universitat Autònoma Barcelona, Bellaterra, ESP); Salazar-Ciudad, Isaac (EvoDevo Helsinki Community, FIN)

C7-02 **The establishment of left-right asymmetry during spiralian development in the serpulid annelid *Pomatoceros lamarcki***
Namigai, Erica (University of Oxford, GBR); Shimeld, Sebastian (University of Oxford, GBR)

C7-03 **Colony symmetry in thecate hydroids (Cnidaria, Hydroidomedusa, Leptomedusae): Transition from radial to bilateral symmetry**
Kosevich, Igor (Lomonosov Moscow State University, RUS)

C7-04 **Evolution of a novel flower trait in the Brassicaceae**
Busch, Andrea (University of Osnabrück, GER); Horn, Stefanie (University of Osnabrück, GER); Zachgo, Sabine (University of Osnabrück, GER)

16.10 – 17.10 **Contributed Session C8:**
Perspectives on Wnt signaling I

ROOM C1

Chair: Reinhard Schröder

C8-01 **A Wnt landscape regulates segment polarity in the annelid *Platynereis***
Balavoine, Guillaume (Institut Jacques Monod, Paris, FRA); Gazave, Eve (Institut Jacques Monod / CNRS, Paris, FRA)

C8-02 **Functional analysis of Wnt signalling in early spider development shows diverse roles in embryonic patterning**
Eibner, Cornelius (Friedrich Schiller University Jena, GER); Pohl, Kerstin (Friedrich Schiller University Jena, GER); Beyerlein, Anna (Friedrich Schiller University Jena, GER); Holzem, Michaela (Friedrich Schiller University Jena, GER); Damen, Wim GM (Friedrich Schiller University Jena, GER)

C8-03 **Characterization of the gene regulatory network for posterior development in the spider *Parasteatoda tepidariorum***
Schoenauer, Anna (Oxford Brookes University, GBR); Schwager, Evelyn (Oxford Brookes University, GBR); Hilbrant, Maarten (Oxford Brookes University, GBR); Damen, Wim G. M. (Friedrich Schiller Universität Jena, GER); McGregor, Alistair P. (Oxford Brookes University, GBR)

C8-04 **Anterior/posterior patterning of the dorsal mesoderm in vertebrates evolved as a novelty from the ancestral chordate mesoderm by a heterotopic shift**
Onai, Takayuki (RIKEN Center for Developmental Biology, Kobe, JPN); Aramaki, Toshihiro (Osaka University, Osaka, JPN); Inomata, Hidehiko (RIKEN Center for Developmental Biology, Kobe, JPN); Hirai, Tamami (RIKEN Center for Developmental Biology, Kobe, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

16.10 – 17.10 **Contributed Session C9:**
Mechanical mechanisms of development

ROOM C2

Chair: Derek Moulton

C9-01 **The 3D Crocs project: Physical mechanisms generate a diversity of cranial scale patterns among Crocodylia**
May, Catherine (University of Geneva, CHE); Milinkovitch, Michel (University of Geneva, CHE)

C9-02 **Epithelial cell shaping in response to mechanical cues is an evolutionary conserved way for sculpting an embryo**
Kraus, Yulia (Lomonosov Moscow State University, RUS); Kremnyov, Stanislav (Lomonosov Moscow State University, RUS); Frank, Uri (National University of Ireland, Galway, IRL)

C9-03 **Comparative tissue dynamics of late embryogenesis in flies and midges**
Fraire-Zamora, Juan J. (Centre de Regulacio Genomica, Barcelona, ESP); Solon, Jerome (Centre de Regulacio Genomica, Barcelona, ESP); Jaeger, Johannes (Centre de Regulacio Genomica, Barcelona, ESP)

C9-04 **Shaping the snapdragon's mouth using a CUP**
Rebocho, Xana (John Innes Centre, Norwich, GBR); Ableу, Katie (John Innes Centre, Norwich, GBR); Bradley, Desmond (John Innes Centre, Norwich, GBR); Copsey, Lucy (John Innes Centre, Norwich, GBR); Bagham, Andrew (University of East Anglia, Norwich, GBR); Coen, Enrico (John Innes Centre, Norwich, GBR)

16.10 – 17.10 **Contributed Session C10:**
Complexity in gene networks and structures

ROOM D

Chair: Andrew Cridge

C10-01 **Constraining and buffering: Regulation and evolution of complex gene networks**
Cridge, Andrew (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL)

C10-02 **Structure, function and development of an extraordinary insect eye**
Buschbeck, Elke (University of Cincinnati, OH, USA); Stahl, Aaron (University of Cincinnati, OH, USA); Cook, Tiffany (Cincinnati Children's Hospital Medical Center, OH, USA)

C10-03 **Evolution of morphological complexity and modularity in the primate skull using anatomical network analysis**
Esteve-Altava, Borja (University of Valencia, Paterna, ESP); Boughner, Julia (University of Saskatchewan, Saskatoon, SK, CAN); Diogo, Rui (Howard University, Washington DC, USA); Rasskin-Gutman, Diego (University of Valencia, Paterna, ESP)

C10-04 **Elastin gene neo-functionalization endows teleost-specific heart component, "bulbus arteriosus", in fish development and evolution**
Moriyama, Yuuta (University of Tokyo, JPN); Takeuchi, Jun K. (University of Tokyo, JPN); Koshiba-Takeuchi, Kauzko (University of Tokyo, JPN)

17.20 – 18.00 Keynote Lecture (K2)

A "Devo-Evo" approach to unravel the hidden logic of cell and tissue polarity in plants and animals

ROOMS C1&2

Veronica Grieneisen
(John Innes Centre, Norwich, GBR)
Chair: Charlie Scutt

18.00 – 20.00 Poster Session 1
Corridors C and ROOM E
(even numbers)

20.30 Reception at Vienna City Hall

Thursday, July 24th

09.00 – 10.40 Symposium S9:

Plant EvoDevo: Linking cross-species genetic and morphological variation

ROOM A

Organizers: John Bowman and Christian Hardtke
Chair: Christian Hardtke

S9-01 **Inverted regulatory logic in hormone pathway interactions shapes different root system types**
Hardtke, Christian (University of Lausanne, CHE)

S9-02 **Genetic determinants of petal number variation between *Arabidopsis thaliana* and *Cardamine hirsuta***
Monniaux, Marie (Max Planck Institute for Plant Breeding Research, Cologne, GER)

S9-03 **A division in PIN-mediated Auxin patterning during organ initiation in grasses**
O'Connor, Devin (Cambridge University, GBR); Runions, Adam (University of Calgary, AB, CAN); Sluis, Aaron (University of California Albany, CA, USA); Bragg, Jennifer (USDA, Albany, CA, USA); Vogel, John (USDA, Albany, CA, USA); Prusinkiewicz, Przemyslaw (University of Calgary, AB, CAN); Hake, Sarah (University of California, Albany, CA, USA)

S9-04 **Towards understanding the genetic basis for diversification of leaf forms**
Tsiantis, Miltos (Max Planck Institute for Plant Breeding Research, Cologne, GER)

09.00 – 10.40 Symposium S10:

What should bioinformatics do for EvoDevo?

ROOM B

Organizers: Günter Plickert, Mark Blaxter, Paula Mabee and Ann Burke
Chair: Paula Mabee

S10-01 **Bioinformatics for Evo Devo: Connecting evolutionary morphology and model organism genetics**
Mabee, Paula (University of South Dakota, Vermillion, SD, USA)

S10-02 **Insights into the evolution and development of planarian regeneration from the genome of the flatworm, *Girardia tigrina***
Kumar, Sujai (University of Oxford, GBR); Kao, Damian (University of Oxford, GBR); Aboobaker, Aziz (University of Oxford, GBR)

S10-03 **From the wet lab to the computer and back: A stage specific RNAseq analysis elucidates the molecular underpinnings and evolution of Hydrozoan development**
Schiffer, Philipp (University of Cologne, GER)

S10-04 **Insights into the evolution of early development of parthenogenetic nematodes by second generation sequencing**
Kraus, Christopher (University of Cologne, GER); Schiffer, Philipp (Universität of Cologne, GER); Kroiher, Michael (University of Cologne, GER); Schierenberg, Einhard (University of Cologne, GER)

09.00 – 10.40 Symposium S11:

Ecological and environmental impacts on the evolution of organismal development I

ROOM C1

Organizers: Chris Lowe, John Willis and Angelika Stollewerk
Chair: Angela Stollewerk

S11-01 **EcoEvoDevo and the origins of morphological complexity in the worker caste in ants**
Rajakumar, Rajendhran (McGill University, Montreal, QC, CAN); Fave, Marie-Julie (McGill University, Montreal, QC, CAN); **Abouheif, Ehab** (McGill University, Montreal, QC, CAN)

S11-02 **Inducible defenses in Daphnia**
Laforsch, Christian (University of Bayreuth, GER)

S11-03 **Genetics of larval mode in the poecilogonous polychaete, *Streblospio benedicti***
Rockman, Matthew (New York University, NY, USA)

S11-04 **Determination of sexual fate in the temperature dependent red-eared slider turtle, *Trachemys scripta elegans***
Capel, Blanche (Duke University, Durham, NC, USA); Czerwinski, Mike (Duke University, Durham, NC, USA); Mork, Lindsey (Duke University, Durham, NC, USA); Looger, Loren (Janelia Farm Research Campus, Ashburn, VA, USA); Natarajan, Anirudh (Duke University, Durham, NC, USA)

09.00 – 10.40 Symposium S12: Developmental basis of quantitative variation

ROOM C2

Organizers: Mihaela Pavlicev and Günter Wagner
Chair: Mihaela Pavlicev

S12-01 **The evolution of pleiotropy in relation to integration, modularity, and individuation**
Cheverud, James M. (Loyola University, Chicago, USA)

S12-02 **Mutational variation in epistatic pleiotropy and the genotype-phenotype map of multi-drug resistance in HIV-1**
Guillaume, Frederic (University of Zurich, CHE)

S12-03 **Highly monotone genotype-phenotype maps emerging from gene regulatory networks**
Gjuvsland, Arne (Norwegian University of Life Sciences, Ås, NOR); Wang, Yunpeng (Norwegian University of Life Sciences, Ås, NOR); Plahte, Erik (Norwegian University of Life Sciences, Ås, NOR); Omholt, Stig W. (Norwegian University of Science and Technology, Trondheim, NOR)

S12-04 **Managing the pleiotropy of "pleiotropic" transcription factor genes**
Wagner, Günter (Yale University, New Haven, CT, USA); Lynch, Vincent (The University of Chicago, IL, USA); Nnamani, Mauris (Yale University, New Haven, CT, USA); Pavlicev, Mihaela (University of Cincinnati, OH, USA)

11.10 – 12.50 Symposium S13: Structural organization in vertebrate dentitions: Molecules, morphology and function

ROOM A

Organizers: Abigail Tucker and Moya Smith
Chair: Moya Smith

S13-01 **Teeth inside and outside the mouth: Topographic relationships in sawshark and sawfish dentitions (Elasmobranchii; Chondrichthyes)**
Welten, Monique (Natural History Museum, London, GBR); Meredith Smith, Moya (King's College London, GBR); Underwood, Charlie (University of London, GBR); Fraser, Gareth (University of Sheffield, GBR); Johanson, Zerina (Natural History Museum, London, GBR)

S13-02 **Mechanical constraints during cusp pattern development in rodent molars**
Renvoise, Elodie (University of Helsinki, FIN); Kavanagh, Kathryn (University of Massachusetts, Dartmouth, MA, USA); Kallonen, Aki (University of Helsinki, FIN); Häkkinen, Teemu (University of Helsinki, FIN); Rice, Ritva (University of Helsinki, FIN); Jernvall, Jukka (University of Helsinki, FIN)

S13-03 **Development and fate of the dental lamina in amniotes**
Buchtova, Marcela (Academy of Sciences of the Czech Republic, Brno, CZE)

S13-04 **What the Myotragus evolutionary lineage tell us about the selective pressures that drive dentition patterning in mammals**
Jordana, Xavier (Institut Català de Paleontologia Miquel Crusafont, Bellaterra, ESP); Moncunill-Solé, Blanca (Institut Català de Paleontologia Miquel Crusafont, Bellaterra, ESP); Köhler, Meike (Institut Català de Paleontologia Miquel Crusafont and ICREA, Bellaterra, ESP)

11.10 – 12.50 Symposium S14: What should bioinformatics do for EvoDevo? II

ROOM B

Organizers: Günter Plickert, Mark Blaxter, Paula Mabee, and Ann Burke
Chairs: Günter Plickert and Mark Blaxter

S14-01 **Petaloidy, polarity and pollination: The evolution of organ morphology networks**
Specht, Chelsea (University of California Berkeley, CA, USA); Yockteng, Roxana (University of California Berkeley, CA, USA); Almeida, Ana M. R. (University of California Berkeley, CA, USA); Pineyro-Nelson, Alma (University of California Berkeley, CA, USA)

S14-02 **Aligning phonemes and genomes to understand the evolution of multicellular organisms**
Donoghue, Philip (University of Bristol, GBR); Deline, Bradley (University of West Georgia, Carrollton, GA, USA); Greenwood, Jennifer (University of Bristol, GBR); Taylor, Richard (University of Bristol, GBR); Hetherington, Alexander (University of Bristol, GBR); Tarver, James (University of Bristol, GBR); Peterson, Kevin (Dartmouth College, Hanover, NH, USA)

S14-03 **Online databases provide critical insights into the evolution of appendage modularity during the fin to limb transition**
Dececchi, Alex (University of South Dakota, Vermillion, SD, USA); Mabee, Paula (University of South Dakota, Vermillion, SD, USA); Marcot, Jonathan (University of Illinois, Urbana, IL, USA); Sears, Karen (University of Illinois, Urbana, IL, USA)

S14-04 **Evolutionally conserved mechanisms of regeneration in chordates: Uncovering pathways active during WBR in *Botrylloides leachi***
Zondag, Lisa (University of Otago, Dunedin, NZL); Rutherford, Kim (University of Ontago, Dunedin, NZL); Wilson, Megan (University of Ontago, Dunedin, NZL)

11.10 – 12.50 Symposium S15:
Ecological and environmental impacts on the evolution of organismal development II

ROOM C1

Organizers: Chris Lowe, John Willis, and Angelika Stollewerk
Chair: Angela Stollewerk

S15-01 **Evolutionary lability of plant epidermal morphology in response to changing interactions with animals**
Glover, Beverley (University of Cambridge, GBR)

S15-02 **New genes, new chemistry and new cells in phenotypic plasticity and the evolution of novelty in nematodes**
Sommer, Ralf (Max-Planck Institute for Developmental Biology, Tübingen, GER)

S15-03 **Understanding strategies used by the *C. elegans* reproductive system to cope with uncertain environments**
Ruvinsky, Ilya (The University of Chicago, IL, USA)

S15-04 **Experimental evolution of multicellularity**
Travisano, Michael (University of Minnesota, Saint Paul, MN, USA); Ratcliff, William (Georgia Institute of Technology, Atlanta, GA, USA)

11.10 – 12.50 Symposium S16:
How does developmental robustness facilitate the evolution of biodiversity?

ROOM C2

Organizers: Rainer Melzer and Günter Theissen
Chairs: Günter Theissen and Rainer Melzer

S16-01 **How do clades explore morphological character space throughout their evolution?**
Wills, Matthew (University of Bath, GBR); Hughes, Martin (Natural History Museum, London, GBR); Gerber, Sylvain (University of Cambridge, GBR); Oyston, Jack (University of Bath, GBR); Wagner, Peter (National Museum of Natural History, Washington DC, USA)

S16-02 **The evolution of canalization and evolvability in changing environments**
Hansen, Thomas F. (University of Oslo, NOR)

S16-03 **Developmental robustness in *C. elegans*: From quantification to mechanisms**
Barkoulas, Michalis (Imperial College, London, GBR); Felix, Marie-Anne (Ecole Normale Superieure, Paris, FRA)

S16-04 **Genetic basis of petal number variation in *Cardamine hirsutas***
Hay, Angela (Max Planck Institute for Plant Breeding Research, Cologne, GER)

14.20 – 16.00 Symposium S17:
EvoDevo as an approach to understanding communication

ROOM A

Organizers: D. Kimbrough Oller and Ulrike Griebel
Chair: D. Kimbrough Oller

S17-01 **Language evolution and change: The impact of modern evolutionary thinking**
Dediu, Dan (Max Planck Institute for Psycholinguistics, Nijmegen, NLD)

S17-02 **The building blocks of language: From molecules to neuronal circuits**
Vernes, Sonja (Max Planck Institute for Psycholinguistics, Nijmegen, NLD)

S17-03 **Computational models of human vocal development and evolution**
Warlaumont, Anne (University of California Merced, CA, USA)

S17-04 **Human vocal development and animal communication in an EvoDevo approach to language**
Oller, D. Kimbrough (University of Memphis, TN, USA); Griebel, Ulrike (University of Memphis, TN, USA)

14.20 – 16.00 Contributed Session C11:
What should Bioinformatics do for EvoDevo?

ROOM B

Chairs: Günter Plickert and Paula Mabee

C11-01 **Phylogenomics of MADS-box genes in flowering plants to identify EvoDevo genes**
Theissen, Guenter (Friedrich Schiller University Jena, GER); Gramzow, Lydia (Friedrich Schiller University Jena, GER)

C11-02 **Illuminating the evolutionary origin of the turtle shell by a comparative tissue-specific transcriptome analysis**
Pascual-Anaya, Juan (RIKEN Center for Developmental Biology, Kobe, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

C11-03 **Blastodermal segmentation in the milkweed bug, *Oncopeltus facsiatus***
Chipman, Ariel (The Hebrew University of Jerusalem, ISR); Stahi, Reut (The Hebrew University, Jerusalem, ISR)

C11-04 **The origins of arthropod innovations: Insights from the non-insect arthropods, the cherry shrimp and rusty millipede**
Kenny, Nathan (The Chinese University of Hong Kong, HKG); Qu, Zhe (The Chinese University of Hong Kong, HKG); Wong, Nicola (The Chinese University of Hong Kong, HKG); Lam, Hon Ming (The Chinese University of Hong Kong, HKG); Chu, Ka Hou (The Chinese University of Hong Kong, HKG); Hui, Jerome (The Chinese University of Hong Kong, HKG)

14.20 – 16.00 Contributed Session C12:
Perspectives on Wnt signaling II

ROOM C1 *Chair:* Wim Damen

C12-01 **Repeated evolution of novel embryonic axis determinants in dipteran insects**
Schmidt-Ott, Urs (The University of Chicago, USA)

C12-02 **Antagonizing Wnt signaling in the Tribolium embryo**
Schröder, Reinhard (University of Rostock, GER); Prühs, Romy (University of Rostock, GER); Sharma, Rahul (University of Rostock, GER); Beermann, Katharina (University of Rostock, GER); Beermann, Anke (Eberhard Karls University of Tübingen, GER)

C12-03 **Wnt-Myc interaction in Hydra stem cell proliferation**
Hobmayer, Bert (University of Innsbruck, AUT); Gufler, Sabine (University of Innsbruck, AUT); Glasauer, Stella (University of Innsbruck, AUT); Bister, Klaus (University of Innsbruck, AUT); Hartl, Markus (University of Innsbruck, AUT)

C12-04 **How regulatory beta-catenin modules impinge on early annelid GRNs**
Schneider, Stephan (Iowa State University, Ames, IA, USA); Pruitt, Margaret (Iowa State University, Ames, IA, USA); Bastin, Benjamin (Iowa State University, Ames, IA, USA); Chou, Hsien-chao (Iowa State University, Ames, IA, USA)

14.20 – 16.00 Contributed Session C13:
Ecological and environmental impacts on the evolution of organismal development I

ROOM C2 *Chair:* Angelika Stollewerk

C13-01 **Combining molecular, developmental, and ecological approaches to understanding the relationship between genotype, phenotype, and the selective environment**
Khila, Abderrahman (Institute of Functional Genomics of Lyon, FRA)

C13-02 **Evolution of the olfactory sensory system in the blind cavefish *Astyanax mexicanus***
Retaux, Sylvie (CNRS, Gif-sur-Yvette, FRA); Bibliowicz, Yoni (CNRS, Gif-sur-Yvette, FRA); Hinaux, Hélène (CNRS, Gif-sur-Yvette, FRA); Blin, Maryline (CNRS, Gif-sur-Yvette, FRA); Alié, Alexandre (CNRS, Gif-sur-Yvette, FRA); Espinasa, Luis (Marist College, Poughkeepsie, NY, USA)

C13-03 **Optogenetics illuminates the neural circuit regulating swimming behavior in the marine annelid *Platynereis dumerilii***
Tosches, Maria Antonietta (European Molecular Biology Laboratory, Heidelberg, GER); Bucher, Daniel (European Molecular Biology Laboratory, Heidelberg, GER); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER)

C13-04 **Genetic an epigenetic bases of abdominal pigmentation plasticity in *Drosophila melanogaster***
Gibert, Jean-Michel (UMR7622 CNRS-UPMC IBPS, Paris, FRA); Mouchel-Vielh, Emmanuèle (UMR7622 CNRS-UPMC IBPS, Paris, FRA); De Castro, Sandra (UMR7622 CNRS-UPMC IBPS, Paris, FRA); Coulpier, Fanny (Genomic Paris Centre, IBENS, Paris, FRA); Le Crom, Stéphane (Genomic Paris Centre, IBENS, Paris, FRA); Peronnet, Frédérique (UMR7622 CNRS-UPMC IBPS, Paris, FRA)

14.20 – 16.00 Contributed Session C14:
Developmental mechanisms underlying evolutionary change II

ROOM D *Chair:* Peter Dearden

C14-01 **A mechanism for reproductive constraint in the honeybee**
Duncan, Elizabeth (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL)

C14-02 **EvoDevo of *Astyanax mexicanus* cavefish: A new time frame and its consequence on the underlying evolutionary mechanisms**
Fumey, Julien (LEGS, UPR9034 CNRS, Gif-sur-Yvette, FRA); Noirot, Céline (Genotoul, INRA, Auzeville, FRA); Hinaux, Hélène (Neurobiology & Development Laboratory UPR3294, Gif-sur-Yvette, FRA); Rétaux, Sylvie (Neurobiology & Development Laboratory UPR3294, Gif-sur-Yvette, FRA); Casane, Didier (LEGS, UPR9034 CNRS, Gif-sur-Yvette, FRA)

C14-03 **Notch signalling is necessary for environmentally-induced transdifferentiation in the sponge, *Amphimedon queenslandica***
Degnan, Bernie (University of Queensland, Brisbane, AUS); Nakanishi, Nagayasu (University of Queensland, Brisbane, AUS); Richards, Gemma (University of Queensland, Brisbane, AUS)

C14-04 **The evolutionary role of microRNA gene regulation in development: Insights from hemichordates**
Gray, Jessica (Harvard Medical School, Boston, MA, USA); Freeman, Jr., Robert (Harvard Medical School, Boston, MA, USA); Gerhart, John (University of California Berkeley, CA, USA); Kirschner, Marc (Harvard Medical School, Boston, MA, USA)

16.30 – 17.00 Contributed Session C15:
EvoDevo as an approach to understanding communication: Modeling, genetics, and developmental research in vocal communication and its neurological underpinnings

ROOM A

Chair: Irene Berra

C15-01 **Endless forms of reward. A combinatorial solution for convergent behavioural traits**
Berra, Irene (University of Amsterdam, NLD, University of Messina, ITA)

C15-02 **Language acquisition as an organic developmental process**
Blasco Máñez, Teresa (The KLI Institute, Klosterneuburg, AUT / University of Oviedo, ESP); Lorenzo, Guillermo (University de Oviedo, ESP); Balari, Sergio (Universitat Autònoma de Barcelona, Bellaterra, ESP)

16.30 – 17.00 Contributed Session C16:
Quo vadis EvoDevo?

ROOM B

Chair: Cassandra Extavour

C16-01 **Remaining questions of the developmental hourglass model**
Irie, Naoki (University of Tokyo, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

C16-02 **Arthropod developmental patterns through time: Is there a decline of diversity?**
Haug, Joachim (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

16.30 – 17.00 Contributed Session C17:
Regeneration EvoDevo

ROOM C1

Chair: Uri Frank

C17-01 **Distinct mechanisms underlie proximal and distal regeneration in the cnidarian, *Hydractinia echinata***
Bradshaw, Brian (National University of Ireland, Galway, IRL; Frank, Uri (National University of Ireland, Galway, IRL)

C17-02 **Homoscleromorpha (Porifera) ectosome regeneration: Morphallaxis and metaplasia**
Ereskovsky, Alexander (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); Borisenko, Ilya (Saint-Petersburg State University, RUS); Gazave, Eve (Institut Jacques Monod, CNRS, Université Paris-Diderot Paris 7, FRA); Renard, Emmanuelle (Aix-Marseille University, FRA); Borchiellini, Carole (Aix-Marseille University, FRA)

16.30 – 17.00 Contributed Session C18:
Ecological and environmental impacts on the evolution of organismal development II

ROOM C2

Chair: Angelika Stollewerk

C18-01 **Regulatory gene networks that shape the development of adaptive phenotypic plasticity in a cichlid fish**
Schneider, Ralf Friedrich (University of Konstanz, GER); Li, Yuanhao (University of Konstanz, GER); Meyer, Axel (University of Konstanz, GER); Gunter, Helen M (University of Konstanz, GER)

C18-02 **Running for life: Developmental and biomechanical constraints on homeotic transformations in mammals**
Galis, Frietson (Naturalis Biodiversity Center, Leiden, NLD); Carrier, David (University of Utah, Salt Lake City, UT, USA); van Alphen, Joris (Groningen University, NLD); Metz, Johan (IIASA, Laxenburg, AUT); Ten Broek, Clara (Naturalis Biodiversity Center, Leiden, NLD)

16.30 – 17.00 Contributed Session C19:
Amphioxus EvoDevo

ROOM D

Chair: Beatriz Albuixech-Crespo

C19-01 **Molecular patterning of amphioxus CNS reveals unexpected evolutionary relationships between midbrain and diencephalon**
Albuixech-Crespo, Beatriz (University of Barcelona, ESP); Irimia, Manuel (University of Barcelona, ESP); Maeso, Ignacio (University of Barcelona, ESP); Sánchez-Arrones, Luisa (CSIC-UAM, Madrid, ESP); Somorjai, Ildiko (University of St Andrews, GBR); Pascual-Anaya, Juan (Riken Center for Developmental Biology, Kobe, JPN); Bovolenta, Paola (CSIC-UAM, Madrid, ESP); Garcia-Fernàndez, Jordi (University of Barcelona, ESP); Ferran, José Luis (University of Murcia, ESP); Puelles, Luis (University of Murcia, ESP)

C19-02 **Functional study of neural induction in the cephalochordate, *Branchiostoma lanceolatum***
Le Pétillon, Yann (UPMC – Laboratoire Arago, Banyuls-sur-Mer, FRA)

17.10 – 17.20 Kowalevsky Medal to
Denis Duboule
(University of Geneva, CHE)

ROOMS C1&2

17.20 – 18.00 Keynote Lecture (K3)
The evolution of key bilaterian traits: Insights from regulatory developmental networks in Cnidaria

ROOMS C1&2

Ulrich Technau
(University of Vienna, AUT)
Chair: Michael Schubert

18.00 – 20.00 Workshop:
Advances in live imaging morphogenesis

ROOM A [& ROOM F]

Chairs: Frederike Alwes and Carsten Wolff
Demonstration

W-01 **High-speed light sheet microscopy and real-time image processing**
Huisken, Jan (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

W-02 **SPIM imaging of spiralian development**
Handberg-Thorsager, Mette (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Tomer, Raju (Stanford University, CA, USA); Amat, Fernando (Janelia Farm Research Campus, Ashburn, VA, USA); Girstmair, Johannes (University College London, GBR); Telford, Max (University College London, GBR); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER); Keller, Philipp (Janelia Farm Research Campus, Ashburn, VA, USA); Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

W-03 **Imaging and quantifying 4D growth patterns during flower development**
Das, Pradeep (ENS Lyon, Lyon, FRA)

W-04 **Multi-level studies of appendage morphogenesis in the crustacean model, *Parhyale hawaiensis***
Pavlopoulos, Anastasios (Howard Hughes Medical Institute, Ashburn, VA, USA); Tinevez, Jean-Yves (Institut Pasteur, Paris, FRA); Pietzsch, Tobias (Max Planck Institute, Dresden, GER); Wolff, Carsten (Humboldt-Universität, Berlin, GER); Tomancak, Pavel (Max Planck Institute, Dresden, GER)

W-05 **Light sheet microscopy as a tool for studying early insect development**
Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

18.00 – 20.00 Poster Session 2
Corridors C and ROOM E
(odd numbers)

Friday, July 25th

09.00 – 10.40 Symposium S18:
EvoDevo of cranial neural crest populations across developmental systems

ROOM A

Organizer: Georgy Koentges
Chair: Georgy Koentges

S18-01 **Characterizing evolutionary conserved regulatory networks in zebrafish craniofacial development**
Eberhart, Johann (University of Texas at Austin, TX, USA); Swartz, M. E. (University of Texas at Austin, TX, USA); Wells, M. B. (University of Texas at Austin, TX, USA); Li, Q. (University of Texas at Austin, TX, USA); Sheehan-Rooney, Kelly (University of Texas at Austin, TX, USA); Rozacky, Jenna (University of Texas at Austin, TX, USA); Dixon, M. J. (University of Manchester, GBR); Vokes, Steven A. (University of Texas at Austin, TX, USA)

S18-02 **Variation in craniofacial derivation and development: Insights from extreme model systems**
Gross, Joshua (University of Cincinnati, OH, USA)

S18-03 **Elaborating a forebrain: Role of the neural crest in vertebrate evolution**
Creuzet, Sophie (Institut de Neurobiologie Alfred Fessard, Gif-sur-Yvette, FRA)

S18-04 **Comparative rhombomeric fate mapping of neural crest and its evolutionary implications**
Koentges, Georgy (University of Warwick, Coventry, GBR)

09.00 – 10.40 Symposium S19:
Quantitative EvoDevo in model and non-model organisms I

ROOM B

Organizers: Benedikt Hallgrimsson, Chris Klingenberg, Philipp Mitteroecker and Ruth Flatscher
Chair: Christian Klingenberg

S19-01 **Morphometrics and the developmental genomics of canalization in craniofacial development**
Hallgrimsson, Benedikt (University of Calgary, AB, CAN); Gonzalez, Paula M. (Universidad Nacional de La Plata, ARG); Mio, Washington (Florida State University, FL, USA); Young, Nathan M. (University of California San Francisco, CA, USA); Percival, Christopher (University of Calgary, AB, CAN); Liberton, Denise (Pennsylvania State University, University Park, PA, USA); Jamniczky, Heather (University of Calgary, AB, CAN); Marcucio, Ralph (University of California San Francisco, CA, USA)

S19-02 **An information theoretic approach to identifying cranial modularity with 3-D morphometric data**
Goswami, Anjali (University College London, GBR); Finarelli, John (University College Dublin, Dublin)

S19-03 **Streptophyta): Quantitative morphometrics at the cellular level**
Neustupa, Jiri (Charles University in Prague, CZE)

S19-04 **Developmental canalization in the vertebrate cranium: A morphometric approach**
Mitteroecker, Philipp (University of Vienna, AUT)

09.00 – 10.40 Symposium S20:
Less is more: Loss of gene functions as a driving force of developmental evolution

ROOM C1

Organizers: Cristian Cañestro and Ingo Braasch
Chairs: Cristian Cañestro and Ingo Braasch

S20-01 **Dynamic gain and loss of genes in animal evolution**
Holland, Peter (University of Oxford, GBR); Quah, Shan (University of Oxford, GBR); Maeso, Ignacio (University of Oxford, GBR); Marletaz, Ferdinand (University of Oxford, GBR); Paps, Jordi (University of Oxford, GBR); Olson, Peter (Natural History Museum, London, GBR); Martin, Kyle (University of Oxford, GBR); Hui, Jerome (The Chinese University of Hong Kong, CHN)

S20-02 **Ohnologs: Why do basal bony fish hold'em but crown groups fold'em?**
Braasch, Ingo (University of Oregon, Eugene, OR, USA); Batzel, Peter (University of Oregon, Eugene, OR, USA); Amores, Angel (University of Oregon, Eugene, OR, USA); Ferrara, Allyse (Nicholls State University, Thibodaux, LA, USA); Fontenot, Quenton (Nicholls State University, Thibodaux, LA, USA); Bobe, Julien (Laboratoire de Physiologie et génomique des poissons, INRA, Rennes, FRA); Postlethwait, John (University of Oregon, Eugene, OR, USA); Guiguen, Yann (Laboratoire de Physiologie et génomique des poissons, INRA, Rennes, FRA)

S20-03 **Revealing cryptic pan-vertebrate gene repertoire in developmental phylome**
Kuraku, Shigehiro (RIKEN Center for Developmental Biology, Kobe, JPN)

S20-04 **A systematic approach to identify gene losses in genome alignments**
Hiller, Michael (Max Planck Institute for Molecular Cell Biology and Genetics (MPI CBG) & Max Planck Institute for the Physics of Complex Systems (MPI PKS), Dresden, GER); Sharma, Virag (MPI CBG & MPI PKS, Dresden, GER); Langer, Bjoern (MPI CBG & MPI PKS, Dresden, GER); Foerster, Leo (MPI CBG & MPI PKS, Dresden, GER); Kiruvale, Pradeep (MPI CBG & MPI PKS, Dresden, GER)

09.00 – 10.40 Symposium S21:
EcoEvoDevo: Symbiosis and epigenetic inheritance

ROOM C2

Organizers: Scott Gilbert and Yoav Soen
Chairs: Yoav Soen and Scott Gilbert

S21-01 **EvoDevo of the holobiont**
Gilbert, Scott (University of Helsinki, FIN)

S21-02 **Experimental evolution of legume endosymbionts**
Masson, Catherine (Laboratory of Plant-Microbe Interactions, INRA, Toulouse, FRA)

S21-03 **Transgenerational inheritance of small RNAs in *C. elegans***
Rechavi, Oded (Tel Aviv University, ISR)

S21-04 **Linking EcoDevo with EvoDevo by non-mendelian inheritance of epigenetic and symbiotic changes**
Soen, Yoav (Weizmann Institute of Science, Rehovot, ISR)

11.10 – 12.25 Contributed Session C20:
EvoDevo of cranial neural crest and dentition

ROOM A

Chairs: Moya Smith and Georgy Koentges

C20-01 **An epithelial stem cell niche and a core gene network regulate continuous tooth regeneration in sharks**
Martin, Kyle (University of Sheffield, GBR); Rasch, Liam (University of Sheffield, GBR); Fraser, Gareth (University of Sheffield, GBR)

C20-02 **Morphogenetics of coordinated tooth and jaw development and evolution in mammals**
Boughner, Julia (University of Saskatchewan, Saskatoon, CAN); Raj, Muhammad (University of Saskatchewan, Saskatoon, CAN); Uppal, Jasmene (University of Saskatchewan, Saskatoon, CAN)

C20-03 **Quantitative modeling of dental stem cell niche evolution and constant change in tooth height over 50 million years**
Mushegyan, Vagan (University of California San Francisco, CA, USA); Eronen, Jussi (University of Helsinki, FIN & Senckenberg Research Institute and Natural History Museum, Frankfurt, GER); Lawing, Michelle (Texas A&M University, College Station, TX, USA); Sharir, Amnon (University of California San Francisco, CA, USA); Janis, Christine (Brown University, Providence, RI, USA); Klein, Ophir D. (University of California San Francisco, CA, USA)

C20-04 **Cellular cartilage predates vertebrates and was coopted by the neural crest during vertebrate head skeleton evolution**
Jandzik, David (University of Colorado at Boulder, CO, USA); Garnett, Aaron T. (University of Colorado at Boulder, CO, USA); Square, Tyler A. (University of Colorado at Boulder, CO, USA); Cattell, Maria V. (University of Colorado at Boulder, CO, USA); Yu, Jr-Kai (Academia Sinica, Taipei, TWN); Medeiros, Daniel M. (University of Colorado at Boulder, CO, USA)

C20-05 **Afferent projections mirror the evolutionary origins of trigeminal sensory neurons**
Butts, Thomas (King's College, London, GBR); Graham, Anthony (King's College, London, GBR)

11.10 – 12.25 **Contributed Session C21:**
Quantitative EvoDevo in model and non-model organisms I

ROOM B

Chair: Philipp Mitteroecker

C21-01 **Adaptive integration in the human pelvis**
Fischer, Barbara (Centre for Ecology and Evolutionary Synthesis, Oslo, NOR); Mitteroecker, Philipp (University of Vienna, AUT)

C21-02 **Grasping flexibility: Evolutionary modularity and developmental origin of carapace integration in Chelonians**
Djurakic, Marko (University of Novi Sad, SRB); Herrel, Anthony (UMR 7179 CNRS/MNHN, Paris, FRA); Jojic, Vida (Institute for Biological Research "Sinisa Stankovic", University of Belgrade, SRB)

C21-03 **Systematic knock-down analysis of the gap gene network in the scuttle fly *Megaselia abdita* reveals quantitative system drift**
Wotton, Karl (Center for Genomic Regulation, Barcelona, ESP); Jimenez-Guri, Eva (Center for Genomic Regulation, Barcelona, ESP); Crombach, Anton (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

C21-04 **Evolution of skull shape in Triturus newts: An ontogenetic and phylogenetic perspective**
Ivanovic, Ana (University of Belgrade, SRB); Cvijanovic, Milena (Institute for Biological Research "Sinisa Stankovic", Belgrade, SRB); Arntzen, Jan W. (Naturalis Biodiversity Center, Leiden, NDL); Zelditch, Miriam (University of Michigan, MI, USA)

C21-05 **Evolution of salamander limbs: Influence of different functional demands of larvae and adults**
Vukov, Tanja (University of Belgrade, SRB); Üzüm, Nazan (Adnan Menderes Universitesi, Aydin, TUR); Urosevic, Aleksandar (University of Belgrade, SRB); Slijepcevic, Maja (University of Belgrade, SRB); Tomasevic Kolarov, Natasa (University of Belgrade, SRB)

11.10 – 12.25 **Contributed Session C22:**
Less is more: Loss of gene functions as a driving force of developmental evolution

ROOM C1

Chairs: Cristian Cañestro and Ingo Braasch

C22-01 **The repertories of developmental transcription factors in sponges were shaped by extensive independent gene loss events**
Fortunato, Sofia (University of Bergen, NOR); Adamski, Marcin (University of Bergen, NOR); Adamska, Maja (University of, NOR)

C22-02 **Lens defects in *Astyanax mexicanus* blind cavefish: Focus on crystallins evolution and function**
Hinaux, Hélène (CNRS Gif-sur-Yvette, FRA); Blin, Maryline (CNRS Gif-sur-Yvette, FRA); Fumey, Julien (CNRS Gif-sur-Yvette, FRA); Legendre, Laurent (CNRS Gif-sur-Yvette, FRA); Casane, Didier (CNRS Gif-sur-Yvette, FRA); Rétaux, Sylvie (CNRS Gif-sur-Yvette, FRA)

C22-03 **Evolution and development of the bifurcated axial skeletal system in the twin-tail goldfish**
Abe, Gembu (Academia Sinica, Yilan, TWN); Lee, Shu-Hua (Academia Sinica, Yilan, TWN); Chang, Mariann (Academia Sinica, Yilan, TWN); Liu, Shih-Chieh (Academia Sinica, Yilan, TWN); Ota, Kinya (Academia Sinica, Yilan, TWN)

C22-04 **A mollusk retinoic acid receptor (RAR) ortholog sheds light on the evolution of ligand binding**
Schubert, Michael (Laboratoire de Biologie du Développement de Villefranche-sur-Mer, FRA)

C22-05 **Darwin's „living fossil" as a new model: Spotted Gar and the genomic basis of vertebrate EvoDevo**
Braasch, Ingo (University of Oregon, Eugene, OR, USA); Batzel, Peter (University of Oregon, Eugene, OR, USA); Loker, Ryan (University of Oregon, Eugene, OR, USA); Amores, Angel (University of Oregon, Eugene, OR, USA); Fontenot, Quenton (Nicholls State University, Thibodaux, LA, USA); Ferrara, Allyse (Nicholls State University, Thibodaux, LA, USA); Postlethwait, John H. (University of Oregon, Eugene, OR, USA)

11.10 – 12.25 **Contributed Session C23:**
EcoEvoDevo: Symbiosis and epigenetic inheritance

ROOM C2

Chairs: Scott Gilbert and Yoav Soen

C23-01 **Development of symbiotic organ: Hox genes regulate development of bacteriome and localization of bacterial symbiont in seed bug *Nysius plebeius***
Matsuura, Yu (Hokkaido University, Sapporo, JPN); Kikuchi, Yoshitomo (Bioproduction Research Insitute, AIST, Hokkaido, Sapporo, JPN); Koga, Ryuichi (Bioproduction Research Institute, AIST, Tsukuba, JPN); Miura, Toru (Hokkaido University, Sapporo, JPN); Fukatsu, Takema (Bioproduction Research Institute, AIST, Tsukuba, JPN)

C23-02 **Ancestral developmental potential facilitates parallel evolution of a novel supersoldier caste in the hyperdiverse ant genus Pheidole**
Rajakumar, Rajendhran (McGill University, Montreal, QC, CAN); San Mauro, Diego (McGill University, Montreal, QC, CAN); Dijkstra, Michiel B. (McGill University, Montreal, QC, CAN); Huang, Ming H. (University of Arizona, Tucson, AZ, USA); Wheeler, Diana E. (University of Arizona, Tucson, AZ, USA); Hiou-Tim, François (McGill University, Montreal, QC, CAN); Khila, Abderrahman (McGill University, Montreal, QC, CAN); Cournoyea, Michael (McGill University, Montreal, QC, CAN); Abouheif, Ehab (McGill University, Montreal, QC, CAN)

C23-03 **Investigating the role of genetic assimilation in an adaptive radiation**
Gunter, Helen (University of Konstanz, GER); Karner, Immanuel (University of Graz, AUT); Schneider, Ralf (University of Konstanz, GER); Sturmbauer, Christian (University of Graz, AUT); Meyer, Axel (University of Konstanz, GER)

C23-04 **Genetic assimilations: How they impact blind variation**
Danchin, Etienne (Evolution & Diversité Biologique, UMR 5174, CNRS, UPS, ENFA, Toulouse, FRA); Pocheville, Arnaud (University of Pittsburgh, PA, USA)

C23-05 **The propensity interpretation of fitness and the Modern Synthesis**
Chiu, Lynn (University of Columbia-Missouri, MO, USA)

11.10 – 12.25 Contributed Session C24: Origin and diversification of regeneration

ROOM D

Chair: Jeremy Brockes

C24-01 **Using sponges to investigate the evolution of stem cell gene regulatory networks**
Revilla-i-Domingo, Roger (Max F. Perutz Laboratories / University of Vienna, AUT); Steudle, Friederike (Max F. Perutz Laboratories / University of Vienna, AUT); Raible, Florian (Max F. Perutz Laboratories / University of Vienna, AUT)

C24-02 **Transcriptional activation of Tgf-beta and Wnt pathways during whole body regeneration in sponges**
Adamski, Marcin (University of Bergen, NOR); Laplante, Mary (University of Bergen, NOR); Liu, Jing (University of Bergen, NOR); Bråte, Jon (University of Oslo, NOR); Leininger, Sven (University of Bergen, NOR); Leon Florian, Luis Anthony (University of Bergen, NOR); Ereskovsky, Alexander (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); Adamska, Maja (University of Bergen, NOR)

C24-03 **Unexpected capacity for the primary body plan regeneration in *Nematostella vectensis* embryos dissociated into single cells**
Kozyreva, Anastasia (Lomonosov Moscow State University, Faculty of Biology, Moscow, RUS); Genikhovich, Grigory (University of Vienna, AUT); Technau, Ulrich (University of Vienna, AUT)

C24-04 **Posterior elongation in the annelid *Platynereis dumerilii* involves stem cells molecularly related to primordial germ cells**
Gazave, Eve (CNRS — Institut Jacques Monod, Paris, FRA); Béhague, Julien (CNRS - Institut Jacques Monod, Paris, FRA); Laplane, Lucie (CNRS — Institut Jacques Monod, Paris, FRA); Demilly, Adrien (CNRS — Institut Jacques Monod, Paris, FRA); Balavoine, Guillaume (CNRS — Institut Jacques Monod, Paris, FRA); Vervoort, Michel (CNRS — Institut Jacques Monod, Paris, FRA)

C24-05 **Early evolution of limb regeneration in tetrapods – evidence from a Palaeozoic amphibian**
Fröbisch, Nadia (Museum für Naturkunde Berlin, GER); Bickelmann, Constanze (Museum für Naturkunde, GER); Witzmann, Florian (Museum für Naturkunde, GER)

14.00 – 15.40 Symposium S22: 'NEPTUNE' ITN: The evolution of sensory systems in the marine environment

ROOM A

Organizers: Andrew Hejnol and Maria Ina Arnone
Chairs: Andrew Hejnol and Maria Ina Arnone

S22-01 **Novel photoreceptors in brachiopod embryos**
Passamaneck, Yale (University of Hawaii, Honolulu, HI, USA); Martindale, Mark (University of Florida, St. Augustine, FL, USA)

S22-02 **Mechanism of phototaxis in *Platynereis* larvae and the origin of visual eyes**
Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

S22-03 **Eye evolution: Common use and independent recruitment of genetic components**
Vopalensky, Pavel (Academy of Sciences of the Czech Republic, Prague, CZE); Pergner, Jiri (Academy of Sciences of the Czech Republic, Prague, CZE); Kozmikova, Iryna (Academy of Sciences of the Czech Republic, Prague, CZE); Kozmik, Zbynek (Academy of Sciences of the Czech Republic, Prague, CZE)

S22-04 **Opsins and the evolution of eyespots in the Acoelomorpha**
Pang, Kevin (University of Bergen, NOR); Hejnol, Andreas (University of Bergen, NOR)

14.00 – 15.40 Symposium S23: Quantitative EvoDevo in model and non-model organisms II

ROOM B

Organizers: Benedikt Hallgrimsson, Chris Klingenberg, Philipp Mitteroecker, and Ruth Flatscher
Chair: Benedikt Hallgrimsson

S23-01 **Yin and yang of EvoDevo: Evolutionary transitions and developmental experiments**
Jernvall, Jukka (University of Helsinki, FIN)

S23-02 **Wnt signalling underlies craniofacial variability in Lake Malawi cichlids**
Parsons, Kevin (Institute of Biodiversity, Animal Health, and Comparative Medicine, Glasgow, GBR); Taylor, Trent (University of Massachusetts Amherst, MA, USA); Powder, Kara (University of Massachusetts Amherst, MA, USA); Albertson, R. Craig (University of Massachusetts Amherst, MA, USA)

S23-03 **Using false flowers to study evolution of modularity and integration**
Armbruster, W. Scott (University of Portsmouth, GBR)

S23-04 **Looking for modularity in morphometric data: Exploratory search without a hypothesis**
Klingenberg, Chris (University of Manchester, GBR)

14.00 – 15.40 Symposium S24: Origin and diversification of regeneration

ROOM C1

Organizer: Florian Raible
Chair: Florian Raible

S24-01 **Taxon-specificity in Salamander limb development and regeneration**
Brockes, Jeremy (University College London, GBR)

S24-02 **Planarians as model system for the evolution of regeneration**
Rink, Jochen (Max Planck Institute for Molecular Cell Biology and Genetics, Dresden, GER)

S24-03 **Germline replacement in the Crustacean, *Parhyale hawaiensis***
Patel, Nipam (University of California Berkeley, CA, USA)

S24-04 **Injury-induced cell death, an evolutionarily-conserved force to drive regeneration?**
Galliot, Brigitte (University of Geneva, CHE); Reiter, Silke (CHE); Wenger, Ivan (CHE); Chara, Osvaldo (CHE); Martinvalet, Denis (CHE); Buzgariu, Wanda (CHE)

14.00 – 15.40 Symposium S25: "Living fossils", myth or reality?

ROOM C2

Organizers: Patrick Laurenti and Didier Casane
Chairs: Didier Casane and Patrick Laurenti

S25-01 **"Living fossils" the facts beyond the myth**
Laurenti, Patrick (CNRS, Université Paris-Diderot, Sorbonne Paris Cité, Gif-sur-Yvette, FRA); Casane, Didier (CNRS, Université Paris-Diderot, Sorbonne Paris Cité, Gif-sur-Yvette, FRA)

S25-02 **Coelacanth and the myth of living fossil**
Dutel, Hugo (RIKEN Center for Developmental Biology, Kobe, Hyogo, JPN)

S25-03 **Evolution and diversity of the Chondrichthyes**
Cuny, Gilles (Natural History Museum of Denmark, Copenhagen, DNK)

S25-04 **Embryology of the hagfish and early evolution of vertebrates**
Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, Hyogo, JPN), Ota, Kinya G. (Marine Research Station, Yilan, TWN), Oisi, Yasuhiro (Max Planck Florida Institute for Neuroscience, Jupiter, FL, USA)

16.10 – 17.10 Contributed Session C25: 'NEPTUNE' ITN: The evolution of sensory systems in the marine environment

ROOM A

Chair: Gaspar Jékely

C25-01 **An ancient neuropeptide regulates both larval settlement and feeding in the marine worm *Platynereis dumerilii***
Williams, Elizabeth (Max Planck Institute for Developmental Biology, Tübingen, GER); Conzelmann, Markus (Max Planck Institute for Developmental Biology, Tübingen, GER); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

C25-02 **Regulation of apical sense organ formation in the sea anemone *Nematostella vectensis* by Frizzled 5/8 and Glypican 4/6**
Bause, Markus (University of Bergen, NOR); Rentzsch, Fabian (University of Bergen, NOR); Leclère, Lucas (Observatoire Océanologique de Villefranche sur Mer, FRA); Sinigaglia, Chiara (Observatoire Océanologique de Villefranche sur Mer, FRA)

C25-03 **Expression of neuropeptides in the brachiopod, *Terebratalia transversa***
Thiel, Daniel (University of Bergen, NOR); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER); Hejnol, Andreas (University of Bergen, NOR)

C25-04 **Evolution and development of photoreceptors in Polyplacophora**
Vöcking, Oliver (University of Bergen, NOR); Hausen, Harald (University of Bergen, NOR)

16.10 – 17.10 Contributed Session C26:
Quantitative EvoDevo in model and non-model organisms II

ROOM B

Chair: Philipp Mitteroecker

C26-01 **A differentially expressed gene network in the head of divergent arctic charr morphs is potentially regulated by Ets-2**
Pashay Ahi, Ehsan (University of Iceland, Reykjavik, ISL); Hristova Kapralova, Kalina (University of Iceland, Reykjavik, ISL); Pálsson, Arnar (University of Iceland, Reykjavik, ISL); Maier, Valerie Helene (University of Iceland, Reykjavik, ISL); Gudbrandsson, Jóhannes (University of Iceland, Reykjavik, ISL); Snorrason, Sigurður (University of Iceland, Reykjavik, ISL); Franzdóttir, Sigrídur R. (University of Iceland, Reykjavik, ISL); Jónsson, Zophonías O. (University of Iceland, Reykjavik, ISL)

C26-02 **Gap domain shifts caused by damped oscillations represent a dynamic fossil of short-germband evolution**
Verd, Berta (Center for Genomic Regulation, Barcelona, ESP); Crombach, Anton (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

C26-03 **Quantification of developmental variation in rainbow trout using geometric morphometric image analysis**
Mayer, Christine (University of Vienna, AUT); Metscher, Brian (University of Vienna, AUT); Müller, Gerd B. (University of Vienna, AUT); Mitteroecker, Philipp (University of Vienna, AUT)

C26-04 **Compartmentalization and spatial complexity of gene expression through development**
Salvador-Martínez, Irepan (University of Helsinki, FIN); Salazar-Ciudad, Isaac (University of Helsinki, FIN)

16.10 – 17.10 Contributed Session C27:
How does developmental robustness facilitate the evolution of biodiversity?

ROOM C1

Chairs: Günter Theissen and Rainer Melzer

C27-01 **The significance of developmental robustness for the biodiversity of life**
Melzer, Rainer (Friedrich Schiller University Jena, GER); Theissen, Günter (Friedrich Schiller University Jena, GER)

C27-02 **A computational approach to the evolution of development under conservative selection**
Zimm, Roland (University of Helsinki, FIN); Salazar-Ciudad, Isaac (University of Helsinki, FIN)

C27-03 **Divergent role of the Hox gene *Antennapedia* in spiders is responsible for the convergent evolution of abdominal limb repression**
Khadjeh, Sara (University of Göttingen, GER); Turetzek, Natascha (University of Göttingen, GER); Prpic-Schäper, Nikola-Michael (University of Göttingen, GER)

C27-04 **Designing a mesodermal molecular toolkit in the marine annelids *Alitta virens* and *Platynereis dumerilii***
Kozin, Vitaly V. (St. Petersburg State University, RUS); Raible, Florian (Max F. Perutz Laboratories, University of Vienna, AUT); Kostyuchenko, Roman P. (St. Petersburg State University, RUS)

16.10 – 17.10 Contributed Session C28:
Uncovering the genomic bases of phenotypic change in the NGS era II

ROOM C2

Chairs: Juan Pascual and Ignacio Maeso

C28-01 **Evolution of the eye transcriptome under constant darkness in Sinocyclocheilus cavefish**
Meng, Fanwei (Chinese Academy of Sciences, Beijing, CHN); Braasch, Ingo (University of Oregon, Eugene, OR, USA); Phillips, Jennifer (University of Oregon, Eugene, OR, USA); Zhang, Chunguang (Chinese Academy of Sciences, Beijing, CHN); Postlethwait, John (University of Oregon, Eugene, OR, USA)

C28-02 **Evolution of the head developmental gene regulatory network in three closely related Drosophila species**
Torres Oliva, Montserrat (Georg August University of Göttingen, GER); Almudi, Isabel (Oxford Brookes University, GBR); Nunes, Maria D. S. (Oxford Brookes University, GBR); McGregor, Alistair P. (Oxford Brookes University, GBR); Posnien, Nico (Georg August University of Göttingen, GER)

C28-03 **Evolutionary innovation by rewiring of gene networks: Origin of sense organs in the vertebrate "new head"**
Riddiford, Nick (National University of Ireland, Galway, IRL); Schlosser, Gerhard (National University of Ireland, Galway, IRL)

C28-04 **Expression and diversification of *CYCLOIDEA* genes in Asteraceae: A case from the highly derived tribe Anthemideae**
Bello Gutiérrez, M. Angélica (Real Jardín Botánico, Madrid, ESP); Cubas, Pilar (Centro Nacional de Biotecnología, Madrid, ESP); Álvarez, Inés (Real Jardín Botánico, Madrid, ESP); Durán, Fátima (Real Jardín Botánico, Madrid, ESP); Sanjuanbenito, Guillermo (Real Jardín Botánico, Madrid, ESP); Fuertes Aguilar, Javier (Real Jardín Botánico, Madrid, ESP)

16.10 – 17.10 Contributed Session C29:
Developmental mechanisms underlying evolutionary change III

ROOM D

Chair: Jack Green

C29-01 **Evolution of the germ cells: Insights from a centipede**
Green, Jack (University of Cambridge, GBR)

C29-02 **DNA methylation and phenotypic plasticity: Is DNA methylation a conserved mechanism for phenotypic plasticity in insects?**
Duncan, Elizabeth (University of Otago, Dunedin, NZL); O'Neill, Meaghan (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL)

C29-03 **Modification of anterior-posterior patterning system toward fin-to-limb transformation: Origin of thumbs and radius**
Onimaru, Koh (Tokyo Institute of Technology, Yokohama, JPN); Kuraku, Shigehiro (RIKEN Center for Developmental Biology, Kobe, JPN); Takagi, Wataru (University of Tokyo, Kashiwa, JPN); Hyodo, Susumu (University of Tokyo, Kashiwa, JPN); Tanaka, Mikiko (Tokyo Institute of Technology, Yokohama, JPN)

C29-04 **Evolution of neurosecretory brain centres: A cell population with an apical-organ-like transcriptional profile pioneers the central nervous system in the centipede, *Strigamia maritima***
Hunnekuhl, Vera (Cambridge University, GBR); Akam, Michael (Cambridge University, GBR)

17.20 – 17.35 Student Poster Prizes

ROOMS C1&2

17.35 – 18.15 Keynote Lecture (K4)
Ancient genes, mesoscale physics, and the origins of animal development

ROOMS C1&2

Stuart Newman
(New York Medical College, USA)
Chair: Frietson Galis

18.15 – 18.20 Conference Closing

ROOMS C1&2

18.20 – 19.10 EED Business Meeting

ROOM C1

19.30 Joint departure for Conference Dinner

Posters

Posters

P-001 **A comparative genomics and transcriptomics approach to the study of *Hox3/zen* gene evolution in insects**
Vargas Jentzsch, Iris (Cologne Biocenter, GER); **Gurska, Daniela** (Cologne Biocenter, GER); Panfilio, Kristen A. (Cologne Biocenter, GER)

P-002 **A comprehensive pipeline for identifiying lncRNAs on the basal-branching chordate Amphioxus**
Herrera, Carlos (University of Barcelona, ESP); Rossell, Ariadna (University of Barcelona, ESP); Burguera, Demian (University of Barcelona, ESP); Irimia, Manuel (University of Barcelona, ESP); Garcia-Fernàndez, Jordi (University of Barcelona, ESP)

P-003 **A developmental model for variation in floral organ number**
Kitazawa, Miho (Osaka University, Toyonaka, JPN); Fujimoto, Koichi (Osaka University, Toyonaka, JPN)

P-004 **A direct transgenic assay to identify highly conserved regulatory sequences acting in Drosophila development**
Schmied, Christopher (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Kalinka, Alexander (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

P-005 **A new self-regulatory model for DV patterning in a basal insect**
Sachs, Lena (University of Cologne, GER); Chen, Yen-Ta (University of Cologne, GER); Roth, Siegfried (University of Cologne, GER)

P-006 **A 3D developmental atlas of Euprymna scolopes (Cephalopoda: Sepiolidae)**
Klimpfinger, Claudia (University of Vienna, AUT); Metscher, Brian (University of Vienna, AUT)

P-007 **Adult prothorax patterning during insect typical metamorphosis**
Hu, Yonggang (Georg August University of Göttingen, GER); Bucher, Gregor (Georg August University of Göttingen, GER)

P-008 **An annelid homolog of the chordate notochord**
Brunet, Thibaut (European Molecular Biology Laboratory, Heidelberg, GER); Lauri, Antonella (European Molecular Biology Laboratory, Heidelberg, GER); Handberg-Thorsager, Mette (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Fischer, Antje H. L. (Harvard University, Cambridge, MA, USA); Simakov, Oleg (European Molecular Biology Laboratory, Heidelberg, GER); Steinmetz, Patrick R. H. (University of Vienna, AUT); Marlow, Heather (European Molecular Biology Laboratory, Heidelberg, GER); Tomer, Raju (Janelia Farm Research Campus, Ashburn, VA, USA); Keller, Philipp J. (Janelia Farm Research Campus, Ashburn, VA, USA); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER)

P-009 **Analysis of embryonic eye development in the spider *Parasteatoda tepiariorum***
Schomburg, Christoph (Georg August University of Göttingen, GER); Schacht, Magdalena (Georg August University of Göttingen, GER); Schneider, Julia (Georg August University of Göttingen, GER); Kirfel, Phillipp (Georg August University of Göttingen, GER); Posnien, Nico (Georg August University of Göttingen, GER); Prpic-Schäper, Nikola-Michael (Georg August University of Göttingen, GER)

P-010 **Anatomical Network Analysis (AnNA) in morphological EvoDevo**
Rasskin-Gutman, Diego (University of Valencia, Paterna, ESP); **Esteve-Altava, Borja** (University of Valencia, Paterna, ESP)

P-011 **Axis determination and pattern formation in the pea aphid: Implications of the expression pattern of conserved developmental genes**
Hsiao, Yi-min (National Taiwan University, Taipei, TWN); Chung, Chen-yo (National Taiwan University, Taipei, TWN); Lu, Hsiao-ling (National Taiwan University, Taipei, TWN); Cook, Charles E. (European Molecular Biology Laboratory — European Bioinformatics Institute, Cambridge , GBR); Chang, Chun-che (National Taiwan University, Taipei, TWN)

P-012 **Axis patterning of the tick, *Boophilus microplus***
Tobias Santos, Vitoria (Universidade Federal do Rio de Janeiro, Macaé, BRA); Monteiro de Barros, Cintia (Universidade Federal do Rio de Janeiro, Macaé, BRA); Logullo, Carlos (Universidade Estadual do Norte Fluminense Darcy Ribeiro, Campos dos Goytacazes, Rio de Janeiro, BRA); Ribeiro, Lupis (Universidade Estadual do Norte Fluminense Darcy Ribeiro, Campos dos Goytacazes, Rio de Janeiro, BRA); Martins Feitosa, Natalia (Universidade Federal do Rio de Janeiro, Macaé, BRA); Campos, Eldo (Universidade Federal do Rio de Janeiro, Macaé, BRA); Marcolla Araujo, Helena (Universidade Federal do Rio de Janeiro, BRA); Fontenele, Marcio (Universidade Federal do Rio de Janeiro, BRA); Nunes da Fonseca, Rodrigo (Universidade Federal do Rio de Janeiro, Macaé, BRA)

P-013 **Bio-cultural ontogenies in Mesoamerica: Data, epistemic issues, and the archaeological roots of EcoEvoDevo and niche construction theory**
Vergara-Silva, Francisco (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX)

P-014 **Biophysical dynamic module for the polarization of auxin efflux carriers PIN-FORMED (PIN)**
Hernández-Hernández, Valeria (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX); Garay, Adriana (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX); Barrio, Rafael (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX); Alvarez-Buylla Roces, Elena (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX); Benitez, Mariana (Universidad Nacional Autonoma de Mexico, Mexico DC, MEX)

P-015 **Brain development of the Hagfish, with reference to the vertebrate brain evolution**
Sugahara, Fumiaki (Hyogo College of Medicine, Nishinomiya, JPN); Oisi, Yasuhiro (RIKEN Center for Developmental Biology (CDB), Kobe, Hyogo, JPN); Pascual-Anaya, Juan (RIKEN CDB, Kobe, Hyogo, JPN); Kuraku, Shigehiro (RIKEN CDB, Kobe, Hyogo, JPN); Aota, Shin-ichi (RIKEN CDB, Kobe, Hyogo, JPN); Adachi, Noritaka (RIKEN CDB, Kobe, Hyogo, JPN); Murakami, Yasunori (Ehime University, Matsuyama, Japan, JPN); Kuratani, Shigeru (RIKEN CDB, Kobe, Hyogo, JPN)

P-016 **Can a subtle change in morphology propel groups of organisms into adaptive radiation?**
Crumière, Antonin (Institute of Functional Genomics, Lyon, FRA); Khila, Abderrahman (Institute of Functional Genomics, Lyon, FRA)

P-017 **Cell and tissue dynamics during *Tribolium castaneum* embryogenesis revealed by versatile fluorescence labelling approaches**
Benton, Matthew A (University of Cologne, GER); Akam, Michael (University of Cambridge, GBR); Pavlopoulos, Anastasios (Howard Hughes Medical Institute Janelia Farm Research Campus, Ashburn, VA, USA)

P-018 **Cellular and molecular analysis of muscles in two Cnidarian species (*Nematostella vectensis*, *Aurelia aurita*)**
Jahnel, Stefan (University of Vienna, AUT); Walzl, Manfred (University of Vienna, AUT); Technau, Ulrich (University of Vienna, AUT)

P-019 **Characterizing the gene regulatory network active in early embryogenesis of the milkweed bug, *Oncopeltus fasciatus***
Novikov, Anastasia (The Hebrew University of Jerusalem, Jerusalem, ISR); Chipman, Ariel (The Hebrew University of Jerusalem, ISR)

P-020 **Cis-regulatory evolution and functional diversification of gene duplicates**
Tanaka, Kohtaro (Instituto Gulbenkian de Ciência, Lisboa, PRT); Hazbun, Alexis (Instituto Gulbenkian de Ciência, Oeiras, PRT); Diekmann, Yoan (Instituto Gulbenkian de Ciência, Oeiras, PRT); Gonzalez, Luís (Instituto Gulbenkian de Ciência, Oeiras, PRT); Vreede, Barbara (Instituto Gulbenkian de Ciência, Oeiras, PRT); Roch, Fernando (Université de Toulouse, FRA); Sucena, Élio (Instituto Gulbenkian de Ciência, Oeiras, PRT)

P-021 **Commissureless regulation of Slit-Robo signalling in insects**
Seeger, Mark (Ohio State University, Columbus, OH, USA)

P-022 **Comparative cephalochordate transcriptomics: Linking genotype and phenotype at the root of chordate evolution**
Benito, Elia (European Molecular Biology Laboratory, Heidelberg, GER); Simakov, Oleg (European Molecular Biology Laboratory, Heidelberg, GER); Larsson, Tomas (European Molecular Biology Laboratory, Heidelberg, GER); Van Dongen, Stijn (University of Cambridge, GBR); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER); Enright, Anton (University of Cambridge, GBR)

P-023 **Comparative expression analyses of homeobox genes in mollusks**
Wollesen, Tim (University of Vienna, AUT); Rodríguez-Monje, Sonia Victoria (University of Vienna, AUT); Fritsch, Martin (University of Vienna, AUT); Todt, Christiane (University Museum of Bergen, NOR); McDougall, Carmel (University of Queensland, Brisbane, AUS); Degnan, Bernard M. (University of Queensland, Brisbane, AUS); Wanninger, Andreas (University of Vienna, AUT)

P-024 **Comparative transcriptomics to study habitat change and adaptive radiation in water striders**
Armisen, David (Institute of Functional Genomics (IGFL), Lyon, FRA); Refki, Peter (IGFL, Lyon, FRA); Khila, Abderrahman (IGFL, Lyon, FRA)

P-025 **Composition of pre-nervous serotonergic signaling system in early embryonic development of sea urchin, clawed frog and mouse**
Nikishin, Denis (Russian Academy of Sciences, Moscow, RUS); Khramova, Yulia (Lomonosov Moscow State University, RUS); Kremnyov, Stanislav (Lomonosov Moscow State University, RUS); Shmukler, Yuri (Russian Academy of Sciences, Moscow, RUS)

P-026 **Conserved core and exchangeable modulators of the BMP signaling**
Genikhovich, Grigory (University of Vienna, AUT)

P-027 Correlative, quantitative, and molecular 3D imaging for Evo-Devo
Metscher, Brian (University of Vienna, AUT)

P-028 CRISPR/Cas9 induced mutations in the Tribolium single-minded gene (Tc-sim) expose functions in ventral midline and growth zone patterning
Rode, Angelika (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Kalb, Katharina (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Helm, Martin (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Musazzi, Dorothea (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Klingler, Martin (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER)

P-029 Descriptive and statistical analysis of colour pattern evolution in two species of lizards
Manukyan, Liana (University of Geneva, CHE); Montandon, Sophie A. (University of Geneva, CHE); Milinkovitch, Michel C. (University of Geneva, CHE)

P-030 Detecting homoplasy in the origin and evolution of adipose fins
Stewart, Thomas (The University of Chicago, IL, USA); Coates, Michael (The University of Chicago, IL, USA)

P-031 Development and evolution of asymmetric male genitalia in the *Drosophila nannoptera* species group
Lang, Michael (Institut Jacques Monod, Paris, FRA); Lemire, Andrew (Janelia Farm Research Campus, Ashburn, VA, USA); Stern, David (Janelia Farm Research Campus, Ashburn, VA, USA); Orgogozo, Virginie (Institut Jacques Monod, Paris, FRA)

P-032 Development and evolution of dentition pattern for spatial order of tooth replacement in the Batoidea (Chondrichthyes)
Underwood, Charlie (Birkbeck College, University of London, GBR); Johanson, Zerina (Natural History Museum, London, GBR); Welten, Monique (Natural History Museum, London, GBR); Rasch, Liam (University of Sheffield, GBR); Fraser, Gareth (University of Sheffield, GBR); Meredith Smith, Moya (King's College London, GBR)

P-033 Development of wing pattern diversity in *Heliconius butterflies*
Hanly, Joe (University of Cambridge, GBR); Wallbank, Richard (University of Cambridge, GBR); Jiggins, Chris (University of Cambridge, GBR)

P-034 Developmental basis of morphological diversity in the large African barbs from the Lake Tana (Ethiopia) species flock
Borisov, Vasily (Russian Academy of Sciences, Moscow, RUS); Shkil, Fedor (Russian Academy of Sciences, Moscow, RUS);

P-035 Developmental pattern in mantis shrimps: Today and in the past
Wiethase, Joris (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Joachim (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

P-036 Developmental plasticity of germline development in the pea aphid *Acyrthosiphon pisum*
Chang, Chun-che (National Taiwan University, Taipei, TWN); Lin, Gee-way (National Taiwan University, Taipei, TWN); Lu, Hsiao-ling (National Taiwan University, Taipei, TWN); Cook, Charles E (European Molecular Biology Laboratory — European Bioinformatics Institute, Cambridge, GBR); Miura, Toru (Graduate School of Environmental Science, Hokkaido University, Sapporo, JPN)

P-037 Developmental studies on the cucullaris muscle in the Mexican Axolotl (*Ambystoma mexicanum*)
Naumann, Benjamin (Friedrich Schiller University of Jena, GER); Olsson, Lennart (Friedrich Schiller University of Jena, GER)

P-038 Differential canonical Wnt and Hh signaling in head and growth zone of a short germ embryo
Oberhofer, Georg (Georg August University of Göttingen, GER); Bucher, Gregor (Georg August University of Göttingen, GER)

P-039 Disentangling oocyte polarity in panoistic ovaries. The role of Capicua in *Blattella germanica*
Elshaer, Nashwa (Pompeu Fabra University, Barcelona, ESP); Piulachs, Maria-Dolors (Pompeu Fabra University, Barcelona, ESP)

P-040 Dissecting the genetic basis and evolution of form using haploid wasps
Cohen, Lorna (University of Illinois at Chicago, IL, USA); Lynch, Jeremy A. (University of Illinois at Chicago, IL, USA)

P-041 Dorsocross in the evolution of insect (extra)embryonic development
Horn, Thorsten (University of Cologne, GER); Panfilio, Kristen A. (University of Cologne, GER)

P-042 **Dual role of the canonical Wnt pathway in endoderm and posterior development in the brachiopod *Terebratalia transversa***
Martín-Durán, José María (University of Bergen, NOR); Vellutini, Bruno C. (University of Bergen, NOR); Hejnol, Andreas (University of Bergen, NOR)

P-043 **Dynamic evolution of Crx-related homeobox loci in mammals: Birth and death from an unstable genomic region**
Maeso, Ignacio (University of Oxford, GBR); Marlétaz, Ferdinand (University of Oxford, GBR); Irimia, Manuel (Center for Genomic Regulation, Barcelona, ESP); Holland, Peter W. H. (University of Oxford, GBR)

P-044 **Early leg development in *Tribolium castaneum*: The inside out phenotype of the new gene Tc-flipflop**
Thümecke, Susanne (Eberhard Karls University of Tübingen, GER); Beermann, Anke (Eberhard Karls University of Tübingen, GER)

P-045 **Effects of artificially induced heterochronies on serial skeletal elements of teleosts**
Shkil, Fedor (Russian Academy of Sciences, Moscow, RUS); Kapitanova, Daria (Russian Academy of Sciences, Moscow, RUS); Smirnov, Sergey (Russian Academy of Sciences, Moscow, RUS)

P-046 **Endoderm out of the head: Pharyngeal origin of cement glands and external gills in bichir**
Minarik, Martin (Charles University in Prague, CZE); Crkvova, Barbora (Charles University in Prague, CZE); Metscher, Brian (University of Vienna, Wien, AUT); Cerny, Robert (Charles University in Prague, CZE)

P-047 **Evaluating if phenotypic classifiers capture genetic and geographical structure in Panther Chameleons (*F. pardalis*)**
Grbic, Djordje (University of Geneva, CHE); Saenko, Suzanne (University of Geneva, CHE); Milinkovitch, Michel (University of Geneva, CHE)

P-048 **Evidence for an Inhibitory cascade in the development of limbs and digits**
Kavanagh, Kathryn (University of Massachusetts Dartmouth, MA, USA)

P-049 **Evo-diversification and evolution of integration: A phylogenetic approach to understand the evolutionary history of morphological traits**
Benitez, Hugo (University of Manchester, GBR); Klingenberg, Chris (University of Manchester, GBR)

P-050 **Evolution of bivalve by the modification of the cleavage pattern**
Hashimoto, Naoki (University of Tsukuba, JPN); Kurita, Yoshihisa (University of Kyusyu, Fukutsu, JPN); Wada, Hiroshi (University of Tsukuba, JPN)

P-051 **Evolution of brachyury: Role in animal development**
Andrikou, Carmen (University of Bergen, NOR); Arnone, Ina (Stazione Zoologica Anton Dohrn, Naples, ITA); Schwaiger, Michaela (University of Vienna, AUT); Technau, Ulrich (University of Vienna, AUT)

P-052 **Evolution of cephalopod eyes by comparative transcriptome analysis of squid and nautilus**
Ogura, Atsushi (Nagahama Institute of Bio-Science and Technoogy, Nagahama, JPN); Shigeno, Shuichi (JAMSTEC, Yokohama, JPN); Yoshida, Masa-aki (National Institute of Genetics, Mishima, JPN)

P-053 **Evolution of early development of lophotrochozoa: Insight from lophotrochozoa specific homeobox genes**
Morino, Yoshiaki (University of Tsukuba, JPN); Hashimoto, Naoki (University of Tsukuba, JPN); Wada, Hiroshi (University of Tsukuba, JPN)

P-054 **Evolution of germ line segregation and *Nanos* regulation in echinoderms**
Swartz, Zachary (Brown University, Providence, RI, USA); Fresques, Tara (Brown University, Providence, RI, USA); Kikuchi, Mani (University of Tokyo, JPN); Wessel, Gary M. (Brown University, Providence, RI, USA)

P-055 **Evolution of metal response element (MRE)-binding transcription factors in three Branchiostoma species**
Materna, Christopher (Roger Williams University, Bristol, RI, USA); Shin, Paul (City University of Hong Kong, Kowloon, HKG); Sorger, Thomas (Roger Williams University, Bristol, RI, USA)

P-056 **Evolution of pancreatic cell types: Insights from the sea urchin *Strongylocentrotus purpuratus***
Perillo, Margherita (Stazione Zoologica Anton Dohrn, Naples, ITA); Arnone, Maria Ina (Stazione Zoologica Anton Dohrn, Naples, ITA)

P-057 **Evolution of placode-derived neurons assessed by cell type-specific transcriptional profiling**
Patthey, Cedric (University of Umeå, SWE); Clifford, Harry (University of Oxford, GBR); Begbie, Jo (University of Oxford, GBR); Shimeld, Sebastian (University of Oxford, GBR)

P-058 Evolution of Rab32/38 subfamily and their role in pigment cell formation in Chordates
Coppola, Ugo (Stazione Zoologica Anton Dohrn, Naples, ITA); Ristoratore, Filomena (Stazione Zoologica Anton Dohrn, Naples, ITA); D'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Naples, ITA)

P-059 Evolution of sex determining systems in the genus Silene
Janousek, Bohuslav (Academy of Sciences of the Czech Republic, Brno, CZE); Slancarova, Veronika (Academy of Sciences of the Czech Republic, Brno, CZE); Zdanska, Jana (Academy of Sciences of the Czech Republic, Brno, CZE); Zluvova, Jitka (Academy of Sciences of the Czech Republic, Brno, AUT); Talianova, Martina Academy of Sciences of the Czech Republic, Brno, CZE); Zschach, Christian (Academy of Sciences of the Czech Republic, Brno, CZE); Siroky, Jiri (Academy of Sciences of the Czech Republic, Brno, CZE); Kovacova, Viera (Academy of Sciences of the Czech Republic, Brno, CZE); Blavet, Hana (Academy of Sciences of the Czech Republic, Brno, CZE); Danihelka, Jiri (Masaryk University, Brno, CZE); Oxelman, Bengt (University of Gothenburg, SWE); Widmer, Alex (ETH Zurich, CHE); Vyskot, Boris (Academy of Sciences of the Czech Republic, Brno, CZE)

P-060 Evolution of the GRN underlying eye differentiation in closely related Drosophila species
Torres-Oliva, Montserrat (Georg August University of Göttingen, GER); Almudí, Isabel (Oxford Brookes University, GBR); Posnien, Nico (Georg August University of Göttingen, GER); McGregor, Alistair P. (Oxford Brookes University, GBR)

P-061 Evolution of the molecular composition of the Nasonia dorsal-ventral patterning GRN
Pers, Daniel (University of Illinois at Chicago, IL, USA)

P-062 Evolutionary conservation of leftward fluid-flow in left-right axis formation
Vick, Philipp (University of Hohenheim, Stuttgart, GER); Schweickert, Axel (University of Hohenheim, Stuttgart, GER); Thumberger, Thomas (University of Heidelberg, GER); Blum, Martin (University of Hohenheim, Stuttgart, GER)

P-063 Evolutionary novelty, a concept still in search of a definition
Racovski, Thibault (Egenis, University of Exeter, GBR)

P-064 Evolutionary origin and diversification of epidermal barrier proteins in amniotes
Strasser, Bettina (Medical University Vienna, AUT); Mlitz, Veronika (Medical University Vienna, AUT); Hermann, Marcela (Medical University Vienna, AUT); Alibardi, Lorenzo (Università di Bologna, ITA); Tschachler, Erwin (Medical University Vienna, AUT); Eckhart, Leopold (Medical University Vienna, AUT)

P-065 Evolution-development congruence in pattern formation dynamics: Bifurcations in gene expressions and regulation of networks structures
Kohsokabe, Takahiro (Graduate School of Arts and Sciences, University of Tokyo, Tokyo, JPN); Kaneko, Kuihiko (University of Tokyo, JPN)

P-066 Evolving modular gene networks in a multicellular context
Calcott, Brett (Johns Hopkins University, Baltimore, MD, USA)

P-067 Exploring developmental cranial integration in great apes
Scott, Nadia (Max Planck Institute for Evolutionary Anthropology, Leipzig, GER); Strauss, Andre (Max Planck Institute for Evolutionary Anthropology, Leipzig, GER); Neubauer, Simon (Max Planck Institute for Evolutionary Anthropology, Leipzig, GER); Hublin, Jean-Jacques (Max Planck Institute for Evolutionary Anthropology, Leipzig, GER); Gunz, Philipp (Max Planck Institute for Evolutionary Anthropology, Leipzig, GER)

P-068 Exploring floral patterning in Arabidopsis with dynamic models
Collaudin, Samuel (ENS de Lyon, FRA); Das, Pradeep (ENS de Lyon, FRA)

P-069 From hair to spine: Development of enlarged and asymmetrical awl hair in the spiny mouse (*Acomys dimidiatus*)
Montandon, Sophie A. (University of Geneva, CHE); Tzika, Athanasia C. (University of Geneva, CHE); Martins, António F. (University of Geneva, CHE); Chopard, Bastien (University of Geneva, CHE); Milinkovitch, Michel C. (University of Geneva, CHE)

P-070 Functional analysis of the FGF ligands FGF8 and Branchless in the Tribolium embryo
Sharma, Rahul (University of Rostock, GER); Beer, Katharina (University of Rostock, GER); Schmöhl, Felix (University of Rostock, GER); Iwanov, Katharina (University of Rostock, GER); Schröder, Reinhard (University of Rostock, GER)

P-071 Functional consequences of lineage-specific duplications: The example of retinoic acid degradation mechanisms in developing Amphioxus
Carvalho, João E. (Laboratoire de Biologie du Développement de Villefranche-sur-Mer (CNRS/UPMC), FRA); Theodosiou, Maria (Institut de Génomique Fonctionnelle de Lyon (CNRS/ENS Lyon), FRA); Chevret, Pascale (Laboratoire de Biométrie et Biologie Evolutive (CNRS/UCBL), Villeurbanne, FRA); Chen, Jie (Institut de Génomique Fonctionnelle de Lyon (CNRS/ENS Lyon), FRA); de Lera, Angel R. (University of Vigo, Pontevedra, ESP); Laudet, Vincent (Institut de Génomique Fonctionnelle de Lyon (CNRS/ENS Lyon), FRA); Schubert, Michael (Laboratoire de Biologie du Développement de Villefranche-sur-Mer (CNRS/UPMC), FRA)

P-072 **Gene expression patterns in salamander limb development and their potential role in the evolution of preaxial polarity**
Triepel, Sandra (Museum für Naturkunde Berlin, GER); Schneider, Igor (Universidade Federal do Para, Belém, BRA); Mitgutsch, Christian (Museum für Naturkunde Berlin, GER); Fröbisch, Nadia (Museum für Naturkunde Berlin, GER)

P-073 **Gene trapping in the amphipod crustacean *Parhyale hawaiensis***
Alwes, Frederike (ENS-Lyon, Institute of Functional Genomics (IGFL), Lyon, FRA); Enjolras, Camille (ENS-Lyon, IGFL, Lyon, FRA); Averof, Michalis (ENS-Lyon, IGFL, Lyon, FRA)

P-074 **Genetic and evolutionary basis of sensory diversity**
Weinberger, Simon (VIB Center for the Biology of Disease, Leuven, BEL); Hassan, Bassem (VIB Center for the Biology of Disease, Leuven, BEL); Ramaekers, Ariane (VIB Center for the Biology of Disease, Leuven, BEL)

P-075 **Genetic basis of the evolution of differences in eye size between *D. simulans* and *D. mauritiana***
Almudi, Isabel (Oxford Brookes University, GBR); Santos Nunes, Daniela (Oxford Brookes University, GBR); Torres, Montserrat (Georg August University of Göttingen, GER); Arif, Saad (Oxford Brookes University, GBR); Posnien, Nico (Georg August University of Göttingen, GER); McGregor, Alistair Peter (Oxford Brookes University, GBR)

P-076 **Within-species variation in the timing of developmental events: Prevalence, heritability and evolutionary implications**
Tills, Oliver (Plymouth University, GBR); Rundle, Simon (Plymouth University, GBR); Spicer, John (Plymouth University, GBR)

P-077 **Genomic analysis of life cycle evolution in Culex mosquitos**
Scobeyeva, Victoria (Moscow State University, RUS); Asgharian, Hosseinali (University of Southern California Los Angeles, CA, USA); Chang, Peter (University of Southern California Los Angeles, CA, USA); Reisen, William (University California Davis, CA, USA); Lysenkov, Sergey (Moscow State University, Moscow, RUS); Nuzhdin, Sergey (University of Southern California Los Angeles, CA, USA)

P-078 **Geometric morphometrical analysis of the evolutionary development of carnassial teeth in extant canids**
Marquez Gonzalez, Paola Andrea (Universidad Nacional de Colombia, Bogota, COL); Muñoz Duran, Joao (Universidad Nacional de Colombia, Bogota, COL)

P-079 **Germline stem cells and cluster formation in the polytrophic meroistic ovary of *Nasonia vitripennis* (Hymenoptera)**
Griebel, Klaus (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Rübsam, Ralph (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER)

P-080 **Gonad regeneration in medusae of the hydrozoan *Clytia hemisphaerica***
Sinigaglia, Chiara (Laboratoire de Biologie du Développement de Villefranche sur Mer UMR7009 CNRS/UPMC Observatoire Océanologique, FRA); Leclère, Lucas (Laboratoire de Biologie du Développement de Villefranche sur Mer UMR7009 CNRS/UPMC Observatoire Océanologique, FRA)

P-081 **Growth vigour of genotypes with impaired and enhanced S-nitrosothiol signaling indiates nitric oxide feedback regulates assimilation in Arabidopsis**
Frungillo, Lucas (University of Campinas, BRA); Skelly, Michael (University of Edinburgh, GBR); Loake, Gary (University of Edinburgh, GBR); Spoel, Steven (University of Edinburgh, GBR); Salgado, Ione (University of Campinas, BRA)

P-082 **Halisarcidae (Demospongiae) ectosome regeneration: Mesenchymal morphogenesis and epimorphosis**
Borisenko, Ilya (Saint-Petersburg State University, RUS); Adamska, Maja (University of Bergen, NOR); Ereskovsky, Alexander (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology, CNRS, University Aix-Marseille, FRA)

P-083 **Heads and tails: To be or not to be?**
Novikova, Elena (Saint-Petersburg State University, RUS); Bakalenko, Nadezhda (Saint-Petersburg State University, RUS); Kulakova, Milana (Saint-Petersburg State University, RUS)

P-084 **Heterochronies in teleost caudal fin evolution: Experimental evidence**
Kapitanova, Daria (Russian Academy of Sciences, Moscow, RUS); Shkil, Fedor (Russian Academy of Sciences, Moscow, RUS)

P-085 **How to build an ectodermal organ? Inferring the minimal gene networks able to generate different types of ectodermal buds**
Marin-Riera, Miquel (Universitat Autònoma de Barcelona, Cerdanyola del Vallès, ESP); Salazar-Ciudad, Isaac (University of Helsinki, FIN)

P-086 **Hyoid first: Heterochronic development of bichir hyoid metamere**
Stundl, Jan (Charles University in Prague, CZE); Crkvova, Barbora (Charles University in Prague, CZE); Cerny, Robert (Charles University in Prague, Prague, CZE)

P-087 **Identification of a preformed germ plasm in the sexual oviparous pea aphid: A non-canonical case in the Hemimetabola**
Lin, Gee-way (National Taiwan University, Taipei, TWN); Cook, Charles E. (European Molecular Biology Laboratory — European Bioinformatics Institute, Cambridge, GBR); Miura, Toru (Graduate School of Environmental Science, Hokkaido University, Sapporo, JPN); Chang, Chun-che (National Taiwan University, Taipei, TWN)

P-088 **Identification of MADS-box genes of the AP1/FUL clade in *Passiflora edulis* (Passifloraceae)**
Scorza, Livia (University of Campinas, BRA); Dornelas, Marcelo (University of Campinas, BRA)

P-089 **Identification of new dorsoventral patterning genes by differential expression analyses in *Tribolium castaneum***
Frey, Nadine (University of Cologne, GER); Stappert, Dominik (University of Cologne, GER); Roth, Siegfried (University of Cologne, GER)

P-090 **Identification of target genes of the terminal system in *Tribolium castaneum* by next-generation-sequencing**
Pridöhl, Fabian (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Weißkopf, Matthias (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Schoppmeier, Michael (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER)

P-091 **Identifying the developmental genetic basis of craniofacial evolution using Threespine Stickleback**
Alligood, Kristin (University of Oregon, Eugene, OR, USA); Kimmel, Charles (University of Oregon, Eugene, OR, USA); Cresko, William (University of Oregon, Eugene, OR, USA)

P-092 **Independent evolution of snakelike morphologies in Squamata: Do lineages with equivalent morphologies share molecular signatures in developmental genes?**
Kohlsdorf, Tiana (University of São Paulo, Ribeirão Preto, BRA); Grizante, Mariana (University of São Paulo, Ribeirão Preto, BRA); Milograna, Sarah (University of São Paulo, Ribeirão Preto, BRA); Singarete, Marina (University of São Paulo, Ribeirão Preto, BRA); Nery, Mariana (University of São Paulo, Ribeirão Preto, BRA); Guimarães, Pedro (Universidade Federal de Uberlândia, Patos de Minas, BRA)

P-093 **Inferring chewing motion and development from adult dental morphology**
Labonne, Gaëlle (UMR CNRS Biogéosciences 6282, Dijon, FRA); Navarro, Nicolas (UMR CNRS Biogéosciences 6282, Dijon, FRA); Laffont, Rémi (UMR CNRS Biogéosciences 6282, Dijon, FRA); Montuire, Sophie (UMR CNRS Biogéosciences 6282, Dijon, FRA)

P-094 **Integrating phylogenetics, ecology and evo-devo to understand the origin of plant species: The role of spur length evolution in speciation of the genus Linaria**
Fernandez-Mazuecos, Mario (University of Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

P-095 **Investigating complex leaf development with the Bladderwort *Utricularia gibba***
Bushell, Claire (John Innes Centre, Norwich, GBR); Lee, Karen (John Innes Centre, Norwich, GBR); Coen, Enrico (John Innes Centre, Norwich, GBR)

P-096 **Is the successional dental lamina initiated in species with one generation of teeth?**
Dosedelova, Hana (University of Veterinary and Pharmaceutical Sciences, Brno, CZE); Glocova, Kristyna (University of Veterinary and Pharmaceutical Sciences, Brno, CZE); Tichy, Frantisek (University of Veterinary and Pharmaceutical Sciences, Brno, CZE); Buchtova, Marcela (Academy of Sciences of the Czech Republic, Brno, CZE)

P-097 **Left-right asymmetric control in hemichordate acorn worm embryos**
Su, Yi-Hsien (Academia Sinica, Taipei, TWN)

P-098 **Localization of beta1 integrin and fibronectin during mussel development**
Maiorova, Mariia (Russian Academy of Sciences, Vladivostok, RUS); Dyachuk, Vyacheslav (Russian Academy of Sciences, Vladivostok, RUS); Odintsova, Nelly (Russian Academy of Sciences, Vladivostok, RUS)

P-099 **Mapping ephippia color variation in *Daphnia magna***
Marcelino, Ana (CEFE, CNRS, Montpellier, FRA); Haag, Christoph (CEFE, CNRS, Montpellier, FRA); Ebert, Dieter (University of Basel, CHE)

P-100 **Mapping the corn snake pigmentation and colour pattern mutations using NGS technology**
Saenko, Suzanne (University of Geneva, CHE); Andersson, Leif (Uppsala University, SWE); Milinkovitch, Michel (Laboratory of Artificial & Natural Evolution, Geneva, CHE)

P-101 **Mechanically gated ion channels during early Xenopus embryogenesis**
Kremnyov, Stanislav (Lomonosov Moscow State University, RUS); Nikishin, Denis (Lomonosov Moscow State University, RUS)

P-102 **Mechanisms of lateral organ laminarization in angiosperms: Zingiberales and beyond**
Almeida, Ana Maria (UC Berkeley San Francisco, CA, USA)

P-103 **Minimal regulatory network predicts the differentiation and plasticity of T CD4+ lymphocytes**
Martínez Sánchez, Mariana (Universidad Nacional Autónoma de México, Mexico City, MEX)

P-104 **Modulation of Platynereis larval behavior by neuropeptides**
Jasek, Sanja (Max Planck Institute for Developmental Biology, Tübingen, GER); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

P-105 **Molecular and cellular differentiation during the early shell field development in *Lymnaea stagnalis***
Hohagen, Jennifer (Georg-August University Göttingen, GER); Jackson, Daniel J. (Georg-August University Göttingen, Göttingen, GER)

P-106 **Morphological integration of sexual dimorphic traits in human skull**
Medialdea, Laura (Universidad Autónoma de Madrid, Spain, Madrid, ESP); Fruciano, Carmelo (University of Konstanz, GER); Romero, Alejandro (University of Alicante, ESP); González, Armando (Universidad Autónoma de Madrid, ESP)

P-107 **Multi-level feedbacks during Tribolium segmentation**
Vroomans, Renske (Utrecht University, NLD); Hogeweg, Paulien (Utrecht University, NLD); ten Tusscher, Kirsten (Utrecht University, NLD)

P-108 **Multiple developmental roles of a tissue-specific alternative splicing factor across deuterostomes: ESRP genes are master regulators of diverse epithelial functions**
Burguera, Demian (University of Barcelona, ESP); Navas, Enrique (University of Barcelona, ESP); Cuomo, Claudia (Stazione Zoologica Anton Dohrn, Naples, ITA); Racioppi, Claudia (Stazione Zoologica Anton Dohrn, Naples, ITA); Esposito, Rosaria (Stazione Zoologica Anton Dohrn, Naples, ITA); Herrera, Carlos (University of Barcelona, ESP); Albuxeich, Beatriz (University of Barcelona, ESP); Andrikou, Carmen (Stazione Zoologica Anton Dohrn, Naples, ITA); D'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Naples, ITA); Spagnuolo, Antonietta (Stazione Zoologica Anton Dohrn, Naples, ITA); Ristoratore, Filomena (Stazione Zoologica Anton Dohrn, Naples, ITA); Arnone, Maria Ina (Stazione Zoologica Anton Dohrn, Naples, ITA); Irimia, Manuel (Center for Genomic Regulation, Barcelona, ESP); Garcia-Fernàndez, Jordi (University of Barcelona, ESP)

P-109 **Mushroom bodies: Homology or not?**
Weber, Melanie (University of Vienna, AUT); Eriksson, Joakim (University of Vienna, AUT)

P-110 **Y-secretase activity is required for apical organ formation in the sea anemone *Nematostella vectensis***
Steger, Julia (University of Bergen, NOR); Richards, Gemma (University of Bergen, NOR); Rentzsch, Fabian (University of Bergen, NOR)

P-111 **Next-generation approaches to understanding the evolution of germline**
Quan, Honghu (University of Illinois at Chicago, IL, USA)

P-112 **Nitric oxide-neuropeptide interaction in the settlement behavior regulation of the marine annelid, *Platynereis dumerilii***
Ueda, Nobuo (Max Planck Institute for Developmental Biology, Tübingen, GER); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

P-113 **NO chordate evolution**
Annona, Giovanni (Stazione Zoologica Anton Dohrn, Naples, ITA); Palumbo, Anna (Stazione Zoologica Anton Dohrn, Naples, ITA); D'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Naples, ITA)

P-114 **Nodal signaling regulates the innate asymmetry of the Amphioxus pharynx**
Soukup, Vladimir (Academy of Sciences of the Czech Republic, Prague, CZE); Kozmik, Zbynek (Academy of Sciences of the Czech Republic, Prague, CZE)

P-115 **Non-canonical dorsoventral patterning in the moth midge *Clogmia albipunctata***
Jimenez-Guri, Eva (Center for Genomic Regulation, Barcelona, ESP); Alcaine, Anna (Center for Genomic Regulation, Barcelona, ESP); Wotton, Karl R. (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

P-116 **Non-invasive long-term fluorescence live imaging of *Tribolium castaneum* embryos**
Strobl, Frederic (Buchmann Institute for Molecular Life Sciences, Frankfurt am Main, GER)

P-117 **Novel beta-glucosidases of the family GH3 could be involved in the development of various animal groups**
Gabrisko, Marek (Slovakian Academy of Sciences, Bratislava, SVK); Janecek, Stefan (Slovakian Academy of Sciences, Bratislava, SVK)

P-118 **Novel mechanism of TCF function in *Ciona intestinalis***
Kari, Willi (University Innsbruck, AUT); Bertrand, Vincent (IBDM, Marseille, FRA); Rothbächer, Ute (University of Innsbruck, AUT)

P-119 Novel mutation in human AMELX gene is associated with defect in amelogenesis
Novakovic, Ivana (University of Belgrade, SRB); Cvetkovic, Dragana (University of Belgrade, SRB); Aleksic-Babic, Kristina (University of Belgrade, SRB); Toljic, Bosko (University of Belgrade, SRB); Dobricic, Valerija (Neurology Clinic CCS, Belgrade, SRB); Milasin, Jelena (University of Belgrade, SRB)

P-120 Origins and regulation of an eutherian novelty: The BGW cluster
Pérez, Enrique (University of Barcelona, ESP); d'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Napoli, ITA); Garcia-Fernàndez, Jordi (University of Barcelona, ESP)

P-121 Origins of modularity in the Soay sheep skull
Damasceno, Elis (University of Manchester, GBR); Klingenberg, Chris (University of Manchester, GBR)

P-122 Osteogenic differentiation of oral mucosal mesenchymal progenitor cells
Dong, Rui (Capital Medical University, Beijing, CHN); Liu, Xiaoliang (School and Hospital of Stomatology, Wuhan, CHN); Ge, Lihua (Capital Medical University, Beiing, CHN); Fan, Mingwen (School and Hospital of Stomatology, Wuhan, CHN)

P-123 Overexpression of human scute homolog genes in Drosophila
Sun, Boyuan (Sichuan Agricultural University, Chengdu, CHN); Simpson, Pat (University of Cambridge, GBR); Mingyao, Yang (Sichuan Agricultural University, Chengdu, CHN)

P-124 Phenotypic divergence among spadefoot toad species reflects accommodation of mechanisms underlying developmental plasticity
Gomez-Mestre, Ivan (Donana Biological Station, Seville, ESP); Kulkarni, Saurabh (Yale University, New Haven, CT, USA); Buchholz, Daniel (University of Cincinnati, OH, USA)

P-125 Phenotypic divergence is triggered by a bidirectional parental dominance in the transcriptomes of sibling orchid allopolyploids
Diehl, Daniel Jacob (University of Vienna, AUT); Paun, Ovidiu (University of Vienna, AUT); Lorenzo Romero, Maria (University of Vienna, AUT); Balao, Francisco (University of Seville, ESP)

P-126 Photoreceptor cell evolution in ambulacrarian larvae, a first contribution
Valero Gracia, Alberto (Stazione Zoologica Anton Dohrn, Naples, ITA); Ullrich-Lüter, Esther (Museum für Naturkunde, Berlin, GER); D'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Napoli, ITA); Delroisse, Jerome (University of Mons, BEL); Arnone, Maria Ina (Stazione Zoologica Anton Dohrn, Napoli, ITA)

P-127 *Pipistrellus pipistrellus* (Chiroptera, Vespertilionidae) postnatal baculum development
Herdina, Anna Nele (University of Vienna, AUT); Plenk Jr., Hanns (Medical University of Vienna, AUT); Benda, Petr (National Museum, Prague, CZE); Lina, Peter H. C. (Naturalis Biodiversity Center, Leiden, NLD); Herzig-Straschil, Barbara (Natural History Museum, Vienna, AUT); Hilgers, Helge (University of Vienna, AUT); Metscher, Brian D. (University of Vienna, AUT)

P-128 Placode size evolution in *Astyanax mexicanus* blind cavefish
Rétaux, Sylvie (CNRS Gif-sur-Yvette, FRA); Hinaux, Hélène (CNRS Gif-sur-Yvette, FRA); Alié, Alexandre (CNRS Gif-sur-Yvette, FRA); Blin, Maryline (CNRS Gif-sur-Yvette, FRA)

P-129 Positive selection drives gene duplications to fixation in Drosophila
Cardoso Moreira, Margarida (University of Lausanne, CHE); Arguello, J. Roman (University of Lausanne, CHE); Grenier, Jennifer K. (Cornell University, Ithaca, NY, USA); Clark, Andrew G. (Cornell University, Ithaca, NY, USA)

P-130 Pro-differentiation state of Deuterostomes´ nervous system in an evolutionary perspective
Anishchenko, Evgeniya (Stazione Zoologica Anton Dohrn, Napoli, ITA); Arnone, Maria Ina (Stazione Zoologica Anton Dohrn, Naples, ITA); D'Aniello, Salvatore (Stazione Zoologica Anton Dohrn, Naples, ITA)

P-131 Quantitative mechanisms explain system drift in the dipteran gap gene network
Crombach, Anton (Center for Genomic Regulation, Barcelona, ESP); Wotton, Karl (Center for Genomic Regulation, Barcelona, ESP); Jimenez-Guri, Eva (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

P-132 Rearing pups with fostering mothers reveals novel parent-of-origin effects in early vocalizations and attachment behavior
Lassi, Glenda (Istituto Italiano di Tecnologia, Genova, ITA); Tucci, Valter (Istituto Italiano di Tecnologia, Genova, ITA)

P-133 **Receptor mediated endocytosis of Wnt**
Mikosch-Wersching, Melanie (Ruprecht Karls University of Heidelberg, GER)

P-134 **Regulation of internode morphogenesis in colonial hydroid *Dynamena pumila* L. (Hydrozoa, Cnidaria)**
Bolshakov, Fedor (Lomonosov Moscow State University, Moscow, RUS); Kosevich, Igor (Lomonosov Moscow State University, Moscow, RUS)

P-135 **Retention of ancestral developmental potential for dentition in the teleost fish (*Astyanax mexicanus*)**
Stock, David W. (University of Colorado at Boulder, CO, USA); Jandzik, David (University of Colorado at Boulder, CO, USA)

P-136 **Revisiting HOX cluster evolution in Nematoda**
Laetsch, Dominik (University of Edinburgh, GBR); Blaxter, Mark (University of Edinburgh, GBR)

P-137 **Role of adaptors Shc, Dos and Drk in Torso and EGFR signaling in Tribolium**
Majumdar, Upalparna (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Klingler, Martin (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER)

P-138 **Role of epigenetic changes in generating non-adaptive genomic variability and evolutionary novelty**
Guerrero-Bosagna, Carlos (Linköping University, SWE)

P-139 **Role of mechano-dependent ion channels in pulsational growth of colonial hydroids**
Nikishin, Denis (Lomonosov Moscow State University, RUS); Kremnyov, Stanislav (Lomonosov Moscow State University, RUS)

P-140 **Roles of retinoic acid signaling in architecting the nervous system of Amphioxus**
Zieger, Elisabeth (Laboratoire de Biologie du Développement de Villefranche-sur-Mer (UMR 7009 ¬ CNRS/UPMC), FRA); Garbarino, Greta (University of Genova, ITA); Candiani, Simona (University of Genova, Genova, ITA); Croce, Jenifer (Laboratoire de Biologie du Développement de Villefranche-sur-Mer (UMR 7009 - CNRS / UPMC), FRA); Schubert, Michael (Laboratoire de Biologie du Développement de Villefranche-sur-Mer (UMR 7009 - CNRS / UPMC), FRA)

P-141 **Seeing eye to eye with the spiders: Differential expression of eye development genes in different eyes of *Cupiennius salei***
Samadi, Leyli (University of Vienna, AUT); Eriksson, Joakim (University of Vienna, AUT)

P-142 **ShapeQTL: Mapping multiple loci for multi-dimensional trait in R**
Navarro, Nicolas (CNRS UMR6282 Biogeosciences, Dijon, FRA)

P-143 **Shavenbaby functions as a segmentation gene in the short germ embryo of Tribolium and may be regulated by mille-pattes**
Ray, Suparna (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Schnellhammer, Irene (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER); Klingler, Martin (Friedrich Alexander University Erlangen-Nuremberg, Erlangen, GER)

P-144 **Snakes and amphisbaenians share molecular signatures in the Conserved Element B, a regulatory fragment for terminal HoxD expression during vertebrate development**
Milograna, Sarah Ribeiro (University of São Paolo, Ribeirão Preto, BRA); Guimarães, Pedro E. M. (Universidade de Uberlândia, Patos de Minas, BRA); Kohlsdorf, Tiana (University of São Paolo, Ribeirão Preto, BRA)

P-145 **Springtails as basal hexapod models for comparative genetic studies**
Konopova, Barbora (University of Cambridge, GBR); Akam, Michael (University of Cambridge, GBR)

P-146 **Steroid-signalling evolution: The Lophotrochozoan ecdysone receptor**
Páscoa, Inês (University of Porto, PRT); Lopes-Marques, Mónica (University of Porto, PRT); Castro, Filipe (University of Porto, PRT); Santos, Miguel (University of Porto, PRT); Ruivo, Raquel (University of Porto, PRT)

P-147 **Structure, function, conservation, and evolution of C2H2 zinc finger transcription factors in arthropods**
Vreede, Barbara (The Hebrew University of Jerusalem, ISR); Stahi, Reut (The Hebrew University of Jerusalem, ISR); Chipman, Ariel (The Hebrew University of Jerusalem, ISR)

P-148 **Super-size me: On the quest for increasing molar size while maintaining shape**
Christensen, Mona (University of Helsinki, FIN); Moustakas-Verho, Jacqueline (Institute of Biotechnology, Helsinki); Jernvall, Jukka (Institute of Biotechnology, Helsinki)

P-149 ***Sycon ciliatum* (Calcarea, Calcaronea) regeneration peculiarities**
Laplante, Mary (University of Bergen, NOR); Adamska, Maja (University of Bergen, NOR); Leininger, Sven (University of Bergen, NOR); Ereskovsky, Alexander (Institut Méditerranéen de Biodiversité et d´Ecologie marine et continentale, CNRS, Aix-Marseille University, FRA)

P-150 **Temporal shift and axis specification during the evolution of early vertebrate development**
Tsikolia, Nikoloz (Georg August University of Göttingen, GER); Stankova, Viktoria (Georg August University of Göttingen, GER); Viebahn, Christoph (Georg August University of Göttingen, GER)

P-151 **Terminal differentiation in reaction-diffusion models**
Häkkinen, Teemu (Institute of Biotechnology, Helsinki); Jernvall, Jukka (Institute of Biotechnology, Helsinki)

P-152 **Testing the role of amniotic marker genes on (extra)embryonic development in the red flour beetle, Tribolium castaneum**
Seibert, Jan (University of Cologne, Cologne, GER); Panfilio, Kristen A. (University of Cologne, GER)

P-153 **The development of palate of the miniature pig**
Du, Juan (Capital Medical University School of Stomatology, Beijing , CHN); Sun, Lindong (Capital Medical University School of Stomatology, Beijing, CHN); Fan, Zhipeng (Capital Medical University School of Stomatology, Beijing, CHN); Wang, Songlin (Capital Medical University School of Stomatology, Beijing, CHN)

P-154 **The echinoderm larval skeleton as a possible model system for experimental evolutionary biology**
Wada, Hiroshi (University of Tsukuba, JPN); Koga, Hiroyuki (University of Tsukuba, JPN); Morino, Yoshiaki (University of Tsukuba, JPN)

P-155 **The effect of floral variation in the field bean (*Vicia faba*) on pollinator behaviour**
Bailes, Emily (University of Cambridge, GBR); Thomas, Jane (National Institute of Agricultural Botany, Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

P-156 **The effect of oxygen deficiency on early ontogenesis of common toad (*Bufo bufo*)**
Dmitrieva, Elena (Lomonosov Moscow State University, RUS)

P-157 **The evolution of floral traits in the Antirrhineae**
Martinez, Cecilia (University of Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

P-158 **The evolution of the RDH10 gene family: Duplication and lineage-specific loss of a novel member**
Ruivo, Raquel (University of Porto, PRT); Lopes-Marques, Mónica (University of Porto, PRT); Castro, João (University of Porto, PRT); Páscoa, Inês (University of Porto, PRT); Freitas, Renata (University of Porto, PRT); Monteiro, Ana (University of Algarve, Faro); Santos, Miguel (University of Porto, PRT); Castro, Filipe (University of Porto, PRT)

P-159 **The evolutionary origin of the vertebrate midbrain**
Suzuki, Daichi (University of Tsukuba, JPN); Murakami, Yasunori (Yasunori Murakami, Ehime University, Matsuyama, JPN); Wada, Hiroshi (University of Tsukuba, JPN)

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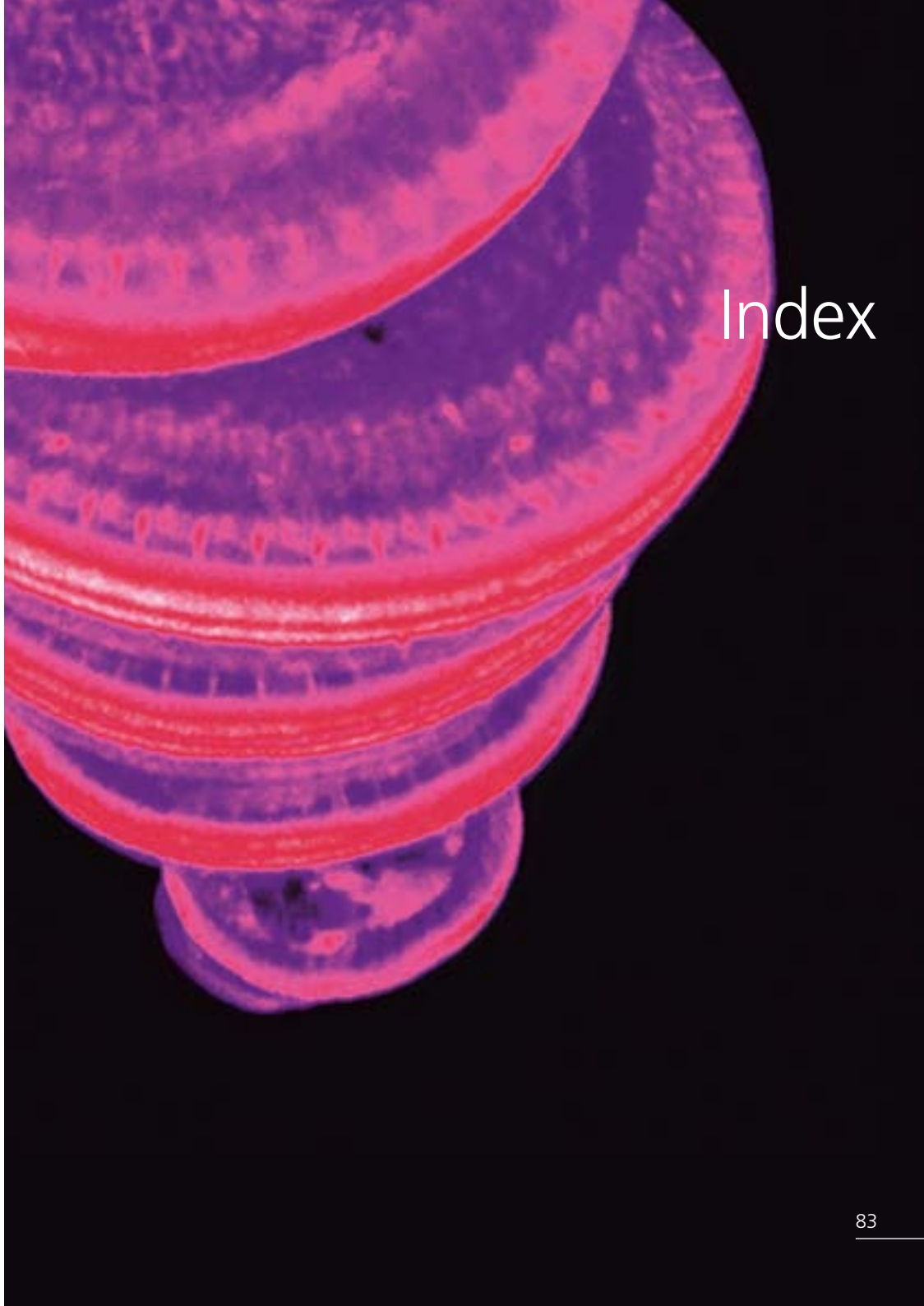

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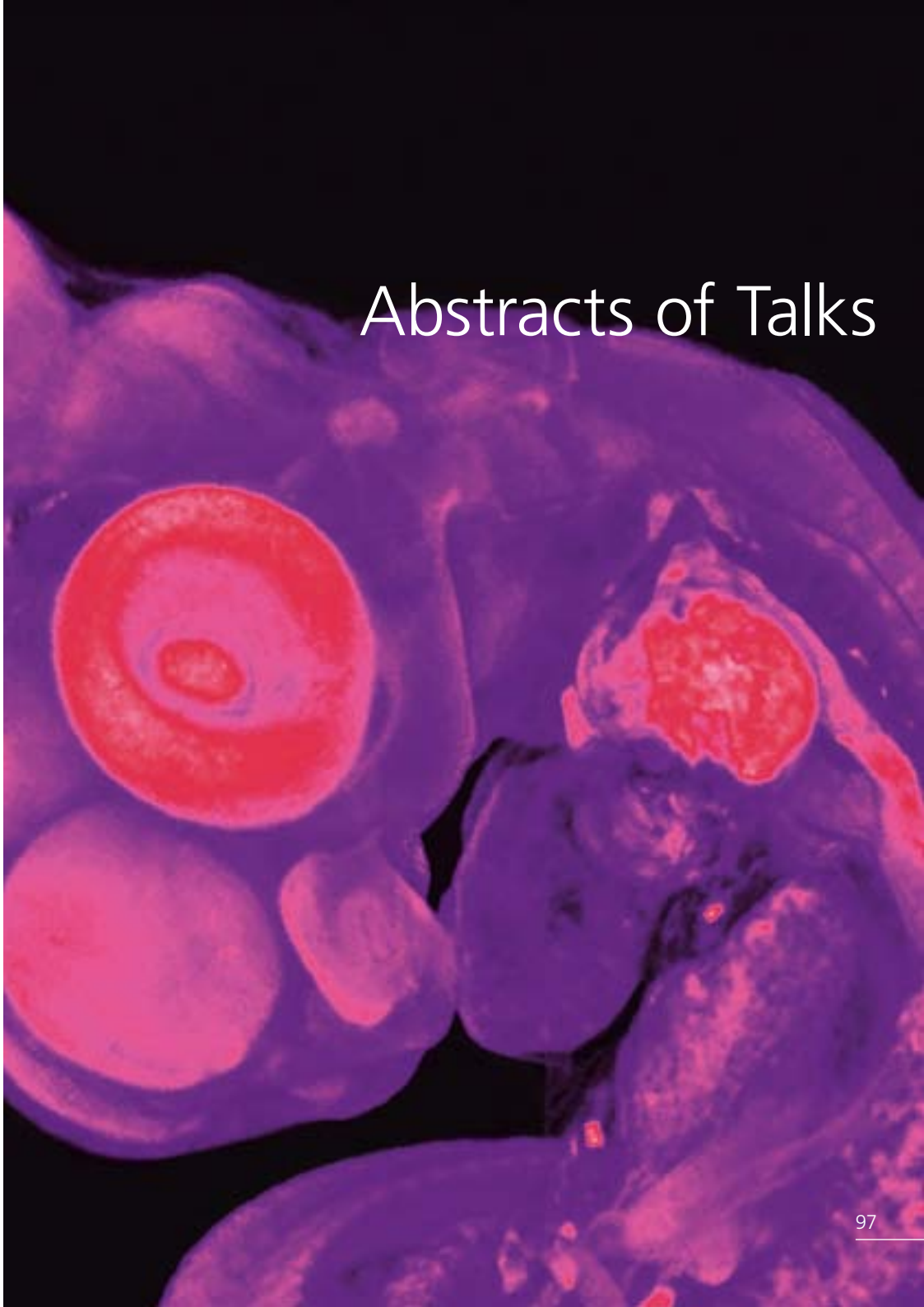
Abstracts of Talks



Abstracts

Tuesday, July 22nd

14.00 – 18.00 Registration

18.00 – 18.20 Opening
ROOMS C1&2
Welcome address by Gerd B. Müller
(President of the EED and Chair of the Local Organizing Committee)

18.20 – 19.00 Keynote Lecture (K1)
Becoming Fully Human
ROOMS C1&2
Jean-Jacques Hublin
(Max Planck Institute for Evolutionary Anthropology, Leipzig, GER)
Chair: Gerd B. Müller

During the last two million years, the increase in behavioural complexity in hominins has been paralleled by a spectacular development of their brain size. Human-accelerated socio-cultural evolution can be viewed as a "niche construction" involving the emergence of unique developmental and demographic patterns. These patterns have directly contributed to the increase in our cognitive abilities. Current fossil evidence suggests that similar selective pressures have been at work in separate hominin lineages following different evolutionary pathways. However, a fully modern life history and cerebral development pattern likely did not appear until rather recently in the course of human evolution. The dramatic expansion of modern humans out of Africa between 100,000 and 50,000 years ago had a major impact on the contemporaneous archaic groups of Eurasia, which were essentially replaced, and ultimately on the whole environment of the planet.

19.00 – 21.00 Welcome Reception at the Venue
sponsored by Springer

Wednesday, July 23rd

09.00 – 10.40 Symposium S1:
Mechanical mechanisms of development I
ROOM A
Organizers: Naomi Nakayama and Annemiek Cornelissen
Chairs: Naomi Nakayama and Annemiek Cornelissen

S1-01 **Patterning under pressure**
Bennett, Malcolm (University of Nottingham, GBR); Goh, Tatsuaki (Kobe University, JPN / University of Nottingham, GBR); Guyomarc'H, Soazig (IRD, UMR DIADE (IRD/UM2), FRA); Fukaki, Hidehiro (Kobe University, JPN); Laplaze, Laurent (IRD, UMR DIADE (IRD/UM2), FRA)

Understanding the mechanisms that control organogenesis remains a central question in plant and animal developmental biology. In contrast to chemical signals such as hormones and peptides, little attention has been paid to mechanical-based patterning mechanisms, yet increasing recent evidence points to a central role for mechanical signals in development in both plants and animals). *Arabidopsis* lateral roots (LRs) originate from a subset of pericycle cells located deep within the primary root. LR formation is initiated when pairs of pericycle cells undergo asymmetric cell division to create two central daughter cells and two larger flanking cells; these cells continue to divide, eventually creating a dome-shaped LR primordium (LRP) that eventually organizes to form a new meristem. New LRP have to emerge through overlying endodermal, cortical, and epidermal tissues that place a biomechanical constraint on new root organs, impacting their morphogenesis. This includes a net-like barrier termed the Casparian Strip (CS) that forms in the overlying endodermal cell layer. The CS appears to impart morphogenetic information by behaving like a "developmental traffic light" that holds back a new LRP (red light) until a mechanical signal triggers the formation of an organising centre and stem cell niche (amber light) just prior to the new organ breaking through the CS and overlying tissues before emerging into the soil (green light). In order to understand how biomechanical and patterning mechanisms interact we have developed a high-resolution time lapse imaging system monitoring LRP development. We will present recent observations revealing several genes and mechanisms controlling the evolution of primordial shape.

S1-02 Getting the mechanics right: The coordination of the lateral expansion of plant stems

Werner, Stephanie (Gregor Mendel Institute, Vienna, AUT); Suer, Stephanie (Gregor Mendel Institute, Vienna, AUT); Wolf, Sebastian (Ruprecht Karl University of Heidelberg, GER); **Greb, Thomas** (Gregor Mendel Institute, Vienna, AUT)

In contrast to animals in which tissue proliferation in adult individuals is often pathological and deleterious, plants have evolved an indefinite growth habit. Especially remarkable in this respect is the thickening of plant stems and roots, as this is a purely postembryonic growth process and the responsible group of stem cells, the cambium, is embedded in highly differentiated tissues. Consequently, lateral plant growth has to integrate and overcome physical constraints implied by surrounding tissues and induce a fundamental change in their properties. This is especially important because cell walls fix the position of plant cells relative to each other and their rigidity is essential for mechanical support of the plant body. Here we explore the process of lateral stem growth in the reference plant *Arabidopsis thaliana* as a paradigm for postembryonic growth processes. We show that hormonal signaling usually involved in mediating general stress response is high in laterally growing stems and that this signaling promotes cambium activity. Being in line with an influence of the cell wall state on lateral stem growth, we identified cbi1, a mutant affected in a gene mediating cell wall rigidity, in a forward mutagenesis screen targeting mutants with altered cambium activity. Furthermore, novel signaling components implicated in cell wall integrity (CWI) influence lateral stem growth. Based on these observations we propose that cell wall-derived and stress-related signals are essential for a coordinated growth of plant organs. By tissue-specific transcriptional profiling of stems at different developmental stages, by ectopic expression of cell wall regulators and by revealing the dynamics of cell wall stiffness during lateral growth at tissue resolution, we are in the process of generating a comprehensive view on a growth process crucial for the dominance of higher plants in continental ecosystems.

S1-03 Cell and tissue mechanics in zebrafish gastrulation

Heisenberg, Carl-Philipp (IST Austria, Klosterneuburg, AUT)

Tissue morphogenesis during embryonic development is brought about by mechanical forces that are generated by the specific biophysical and motility properties of its constituent cells. It has also been suggested that embryonic tissues behave like immiscible liquids with a given surface tension and that differences in surface tension between tissues determine their spatial configuration during embryogenesis. To understand how single cell biophysical and motility properties regulate tissue surface tension and how tissue surface tension controls tissue organization in development, we are studying the specific function of germ layer progenitor cell adhesion, cell cortex tension and motility in determining germ layer organization during zebrafish gastrulation. We found that the combinatorial activity of progenitor cell adhesion, cortex tension and motility determines germ layer tissue surface tension and that differences in germ layer tissue surface tension influence germ layer organization during gastrulation. We will discuss these findings in the light of different hypotheses explaining how single cell biophysical properties determine tissue morphogenesis in development.

S1-04 Sequential rings of cells turn into sequential body parts and sequential evolution by physical pattern formation

Fleury, Vincent (Laboratoire MSC/UMR7057, Paris, FRA)

In his celebrated book, *The Origin of Species* (1859), Charles Darwin states that there should be an ancestor of all vertebrates. It is called the "archetype". However, in the same opus, Darwin states that the "archetype" is an *ideal primitive form*. Being the *primitive form* or *ancestor* of all vertebrates, or being an *ideal form* is not the same thing. Since then, the progress in genetics of development has rendered the concept of "archetype" seemingly obsolete, and even useless. Nevertheless, there might be a need for a mechanism to make new body plans that, by definition, should escape incremental evolution. Physical laws provide such a mechanism, which gives both an *ancestor*, and an *ideal form*. Indeed, recent progress in the physics of morphogenesis has brought back fundamental laws into the problem of embryogenesis. Unexpectedly, a general mechanical law can be put forward, for making the archetype of a vertebrate from a reference state that is a circular plate or a shell. Time-lapse recording at cell-resolution level of early vertebrate (chicken) development shows that the formation of the early embryo occurs by visco-elastic buckling, with folds being locked at boundaries of elastic contrasts. The initial reference state of the chicken embryo is a disc with concentric rings of cells, inherited from the time arrow of cell divisions. By day two of development, mesoderm traction pulls on the blastula and makes it fold. The folds are locked to the boundaries between cell domains, at lines of elastic discontinuities. The traction generates a bilateral animal form and it segregates the cellular territories by the same token. The next rings in the chicken blastula lock one fold forming the amnios and, the next ring forms the yolk sac. Therefore, some aspects in evolution are constrained by the laws of visco-elasticity, and may be generic, hence independent of the exact molecular pathways exerting the forces.

09.00 – 10.40 Symposium S2: EvoDevo of colour

ROOM B

Organizer: Beverley Glover
Chair: Beverley Glover

S2-01 Under the rainbow: Understanding how plants build microscopic structures to produce iridescence

Moyroud, Edwige (University of Cambridge, GBR); Vignolini, Silvia (University of Cambridge, GBR); Rudall, Paula (Royal Botanic Gardens Kew, London, GBR); Steiner, Ullrich (University of Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

Flowering plants also produce microscopic structures to colour their fruits and flowers and insect pollinators can use petal iridescence to detect flowers more efficiently. The physical mechanism responsible for this effect is a surface diffraction grating formed by ordered striations on flat petal cells (like that produced by the data grooves on a CD). The particular amplitude and frequency of the striations cause interference, giving rise to an angular colour variation. These striations are part of the cuticle, a hydrophobic layer consisting of a polymer matrix that incorporates waxes and covers the surface of all plant organs. We aim to understand how an optically accurate diffraction grating can develop on the surface of a flower petal by coupling a range of molecular and cell biology tools with optical analysis and behavioural ecology. As a starting point, we developed a protocol to efficiently transform an iridescent species, *Hibiscus trionum*. We are now using this new model to determine if mechanical buckling of the cuticle could explain the formation of ordered striations on the petal epidermis.

S2-02 Plant genes that alter pollinator preference

Sheehan, Hester (University of Berne, CHE); Klahre, Ulrich (University of Berne, CHE); Dell'Olivo, Alexandre (University of Berne, CHE); Moser, Michel (University of Berne, CHE); Hermann, Katrin (University of Berne, CHE); Esfeld, Korinna (University of Berne, CHE); Summers, Holly (University of Berne, CHE); Caze, Ana Luiza (Universidade Federal do Rio Grande do Sul, Porto Alegre, BRA); Kuhlemeier, Cris (University of Berne, CHE)

Closely related plant species can display dramatically different floral traits and thus can attract different pollinators. We want to understand the concerted molecular and genetic changes that underlie the evolution of these differences. To do this, we chose the *Petunia* genus because of its long history of biochemical and genetic research on petal colour and other relevant traits. The majority of *Petunia* species such as *P. inflata* have purple flowers and display the ancestral pollination syndrome of bee pollination. A clade of closely related derived species show different pollination syndromes. This includes *P. axillaris*, which has white flowers that are ultraviolet (UV)-absorbing and attracts nocturnal moths, and its descendent, *P. exserta*, which has red, UV-reflecting flowers that are typical of hummingbird pollination. The differences in visible and UV floral colour between the Petunia species are due to differing amounts of pigments, anthocyanins and flavonols, respectively.

The evolution of the white-flowered *P. axillaris* from purple-flowered *P. inflata* involved the loss of anthocyanins in floral tissue. This is due to the loss of functionality of a R2R3-MYB transcription factor, AN2, which regulates structural genes in the anthocyanin pathway. In wild accessions of *P. axillaris*, multiple independent loss-of-function mutations have occurred, indicating that loss of colour has happened repeatedly. Transgenic introduction of a functional AN2 into *P. axillaris* altered pollinator preference, demonstrating the importance of a change in a single gene and single trait. Whilst *P. axillaris* has a low level of anthocyanins in its floral tissue, it has a high level of UV-absorbing flavonols. In contrast, the red-flowered *P. exserta* has a low level of these flavonols. QTL analysis combined with transposon tagging and bioinformatic analysis has revealed another R2R3-MYB responsible for the reduction of flavonols in *P. exserta*. The activity of this gene appears to control a trade-off between flavonols and anthocyanins by an as-yet unknown mechanism. Work is underway to study the segregation of this gene in hybrid populations in the natural habitat.

S2-03 Evolution of a novel pigmentation pattern through regulatory rewiring

Martins, Talline (Duke University, Durham, NC, USA); Rausher, Mark (Duke University, Durham, NC, USA)

Petal spots are one of the most common patterns of pigmentation seen in flowers, yet our knowledge of how these patterns form remains limited. In *Clarkia gracilis* (Onagraceae), each petal has a large red-purple spot that contrasts against a light pink background. To identify the spot-determining gene, we employed whole transcriptome sequencing of petals of two *C. gracilis* subspecies that differ in their spot morphologies, coupled to differential expression analyses. This approach uncovered genes that are upregulated in spots relative to unspotted sections of the petals. A SNP in one of these genes, a *R2R3Myb* transcription factor, co-segregates perfectly with spot position. Furthermore, the *R2R3Myb* is only expressed in sections of the petals where spots are formed, suggesting that this transcription factor determines where spots form. Further supporting its role in spot development, in one subspecies of C. gracilis that lacks spots, this *R2R3Myb* has a deletion that results in a premature stop codon. *R2R3Myb* transcription factors are known regulators of anthocyanin pigment

biosynthesis in plants. However, in *C. gracilis*, this *R2R3Myb* appears to have been connected to a novel set of regulators expressed in distinct areas of the petal, leading to the evolution of a novel trait.

S2-04 How chameleons change colour

Milinkovitch, Michel (University of Geneva, CHE); Teyssier, Jeremie (University of Geneva, CHE); Saenko, Suzanne (University of Geneva, CHE); van der Marel, Dirk (University of Geneva, CHE)

Many chameleons, and panther chameleons (*Furcifer pardalis*) in particular, have the remarkable ability to exhibit complex and rapid color changes during social interactions such as male contests or courtship. Combining microscopy, videography, spectroscopy and optical modeling, we show that chameleons shift color through physical mechanisms unrelated to dispersion/aggregation of pigment-containing organelles within dermal chromatophores. We show how these remarkable processes allow many species of chameleons to combine efficient camouflage with spectacular display and resistance to extreme sunlight exposure.

09.00 – 10.40 Symposium S3: Uncovering the genomic bases of phenotypic change in the NGS era

ROOM C1

Organizers: Manuel Irimia, Ignacio Maeso, Juan Pascual-Anaya
Chairs: Manuel Irimia and Ignacio Maeso

S3-01 NG sequencing technology and comparative genomics of non-model systems

Hejnol, Andreas (University of Bergen, NOR); Ryan, Joe F. (University of Bergen, NOR); Vellutini, Bruno C. (University of Bergen, NOR); Martín-Durán, José María (University of Bergen, NOR); Pang, Kevin (University of Bergen, NOR); Børve, Aina (University of Bergen, NOR)

Progress in next-generation sequencing technologies and bioinformatics facilitate the sequencing, assembly, annotation and analyses of novel genomes and transcriptomes. This provides novel insights into genome evolution and accelerates the implementation of molecular methods to non-model systems. Organismal characteristics — such as limited access to material and large genome sizes — restrict the applicability of several of the newest technologies to new species. The information gained by broadening the taxon sampling across the animal tree of life however, overcomes the cost of working with draft genomes alone. Using draft genomes, identification of gene complement and rare genomic changes can help in the placement of animal taxa in the animal phylogeny and contributes to insights about the base of phenotypic change. The genomic information facilitates the establishment of functional methods for developmental studies. Here we will present strategy, experience and results of the sequencing of diverse animal taxa such as acoelomorphs, brachiopods and other spiralians.

S3-02 Mechanisms of gene regulatory evolution during the origin of mammalian pregnancy

Lynch, Vincent J. (The University of Chicago , IL, USA)

How morphological innovations originate is an enduring question in biology. While it's clear that gene regulatory evolution is ultimately responsible for phenotypic differences between species, the molecular mechanisms that underlie the evolution of innovations and major morphological transitions are obscure. Here we use mRNA-Seq to compare gene expression in the pregnant uterus across the major mammalian lineages, including eight placental mammals, two marsupials, and the egg-laying platypus as well as a chicken. We show that thousands of genes were recruited into uterine expression during the evolution of pregnancy, including cell surface proteins that establish implantation of the mammalian embryo (LAMA1/LAMB1/LAMC1), essential signaling pathways that mediate maternal-fetal communication (cAMP, IL11, LIF), and gene regulatory networks that regulate hormone responses to pregnancy (HAND2, AHR). Using complementary experimental methods we show that the transcription factors HAND2 and AHR were recruited into uterine expression through the domestication of placental-mammalian specific transposons into repressor and insulator elements, respectively. Finally we use ChIP-Seq data to demonstrate that >13,300 H3K4me3 marked regulatory elements, ~59% of H3K4me3 peaks, in human uterine cells are derived from transposons indicating that the regulatory landscape of the mammalian genome has been dramatically reorganized by transposons.

S3-03 Understanding extreme non-coding conservation

Lenhard, Boris (Imperial College London, GBR)

Highly conserved non-coding elements (HCNEs) are among the least understood features of metazoan genomes. While their association with transcriptional regulation genes involved in multicellular processes is well established, none of their demonstrated biological functions or combination thereof are able to explain the observed levels and evolutionary depth of their sequence-level conservation.

A general genomic feature of HCNEs is that they do not appear in

isolation but form clusters that span up to megabase regions around their target genes. The regions spanned by this clusters are called genomic regulatory blocks (GRBs). We show that HCNEs in a GRB exhibit coordinated ensemble properties, the most notable of which is the coordinated rate of turnover that differs across different GRBs. By analysing GRB properties and turnover around orthologous genes using multiple reference genomes, we show that the GRBs that appear to have originated from multiple waves of innovation can often be better explained by different turnover rates, and that the appearance of HCNEs around some of GRB target genes likely preceded the root of Metazoa. Finally, we present several computational tools for the analysis and visualisation of key properties of HCNEs and GRBs locus- and genome-wide.

S3-04 Big mice on small islands: The origin and evolution of the Faroese house mouse

Chan, Frank (Friedrich Miescher Laboratory of the Max Planck Society, Tübingen, GER)

Island gigantism has evolved repeatedly in birds and many mammals. Yet little is known about the genetics underlying this ecological rule. We have collected giant wild mice from the Faroe Islands, which are among the largest wild mice in the world and are often 50% heavier than continental mice. In addition, the Faroese mice exhibit significant directional asymmetry in skull development. I will first discuss the origin of these mice, which turns out to be an inter-subspecific hybrid between *Mus musculus domesticus* and *Mus musculus musculus*. We have retraced its evolutionary origin to the early days of colonization of the Faroe Islands. Then I will discuss our on-going efforts to uncover the genetic underpinnings of their large body size. From our laboratory colony we have established a large (>800) F2 intercross panel with a reference-inbred line (SM/J for "small"). For each individual we have gathered a wealth of phenotypes, including growth curves from biweekly measurements, organ weight and plasma growth hormone levels. To dissect the genetic basis of body size variation, we have generated a high-resolution genetic map from our intercross panel (>2 markers per cM) using restriction-site associated DNA sequencing (RADseq). The rich set of genotype–phenotype data allows us to generate a detailed genetic blueprint for body size in a wild mouse population. I will discuss preliminary results arising from this mapping panel. In addition I will discuss pilot results in applying an automated atlas-based image registration pipeline on whole-body 3-dimensional X-ray reconstructions collected from these mice. Applied across the whole panel we may be able to test the genetic validity of various ecological rules regarding climate adaptations like Allen's Rule, Bergmann's Rule or the Island Rule for the first time. The large size of the intercross panel and the wealth of phenotyping information make this a unique wild mouse genetic resource to investigate the evolutionary genetics of complex traits in evolution as well as genetic mechanisms underlying island gigantism.

09.00 – 10.40 Symposium S4: Extended Evolutionary Synthesis

ROOM C2

Organizers: Gerd B. Müller and Werner Callebaut
Chair: Gerd B. Müller

S4-01 Niche construction, developmental bias and the Extended Evolutionary Synthesis

Laland, Kevin (University of St Andrews, GBR); **Uller, Tobias** (University of Oxford, GBR)

"Niche construction" refers to the process whereby the metabolism, activities, and choices of organisms modify or stabilize environmental states, and thereby affect selection acting on themselves and other species. Organisms can modify selective environments externally, for instance, by constructing nests, burrows, mounds, selecting habitat and essential resources, relocating in space (e.g., migration), and leaving ecological legacies for future generations ("ecological inheritance"). A body of formal evolutionary theory has shown that niche construction can strongly affect evolutionary dynamics. The evolutionary significance of niche construction stems from that fact that (1) organisms modify environmental states in *nonrandom* ways, thereby imposing a systematic bias on the selection pressures they generate, and (2) ecological inheritance affects the evolutionary dynamics of descendants, and contributes to the cross-generational stability of environmental conditions that facilitates adaptive evolution. Advocates of niche construction theory have argued that niche construction should be recognized as an evolutionary process (Odling-Smee et al. 2003). In contrast, standard evolutionary theory treats the environment as a "background condition", and restricts evolutionary processes to processes that directly modify gene frequencies. There are parallels between niche construction and developmental bias: niche construction operates like an externally expressed developmental bias to channel selection and impose patterning on evolution. The extended evolutionary synthesis (EES) offers an alternative way of conceptualizing evolution, with its own set of assumptions and processes. Within the EES framework, developmental bias and niche construction would be recognized as evolutionary processes that impose direction on evolution by biasing the action of selection, with developmental plasticity an important source of such bias.

S4-02 An evolutionary view of brain development and dynamics and its theoretical consequences

Szathmáry, Eörs (Parmenides Foundation, Pullach, GER)

Evolutionary dynamics is a general mechanism through which nature develops and maintains highly complex adaptive systems. I shall review research in how far this mechanism can also explain the human brain's remarkable development and functioning. There is new neurophysiological evidence on structural plasticity as well as new models of learning that show how hierarchical representations of complex actions might evolve, mutate and recombine in the brain. There are also new models of adaptive language processing and problem solving that provide interesting windows on how evolutionary dynamics could be relevant for explaining higher-order mental functions. All this raises many fascinating open questions and exciting challenges that we should tackle now in order to understand the full potential of evolutionary neurodynamics to explain our uniquely human capabilities.

S4-03 A new vision of heredity to build an inclusive Evolutionary Synthesis

Danchin, Etienne (CNRS, Toulouse, FRA)

Many biologists are calling for an "extended evolutionary synthesis" that would "modernize the modern synthesis" of evolution. Biological information is typically considered as being transmitted across generations by the DNA sequence alone, but accumulating evidence indicates that both genetic and non-genetic inheritance and their interactions have important effects on evolutionary outcomes. I will briefly review some of the evidence for such effects of epigenetic and cultural inheritance. I will then integrate all forms of inheritance into a single general diagram in order to show how deeply nongenetic inheritance is likely to impact evolutionary processes. Finally, these issues have major implications for diverse domains, including medicine and human sciences where they may profoundly affect scientific research strategies. For instance, non-genetic inheritance may explain a significant part of one of the major enigmas of current molecular biology, namely the case of the missing heritability, which concerns many human supposedly genetic disorders. Jointly with the accruing evidence for nongenetic inheritance, the missing heritability suggests that we should abandon the current genocentric framework of inheritance and adopt a broader view including nongenetic inheritance in order to build the long sought "inclusive Evolutionary Synthesis".

S4-04 The role of EvoDevo in extending the Evolutionary Synthesis

Callebaut, Werner (The KLI Institute, Klosterneuburg, AUT); Müller, Gerd B. (University of Vienna, AUT)

Since the time of the Modern Synthesis of the 1930s and 1940s — the major integrative project in biology hitherto — the biosciences have made diverse significant advances, and today's evolutionary biology operates with numerous concepts, models, and theories that were not part of the original synthesis. Since the retrospective subsumption of these novel accomplishments under the well-defined Synthesis framework is impossible, propositions for a new and extended Evolutionary Synthesis are on the rise. One characteristic feature of this revised framework is the shift from the population-genetic emphasis, favored by traditionalists, towards a multi-level, causal-mechanistic explanation of situated phenotypic complexity. The integration, by EvoDevo, of organizing relations between genes, cells, and tissues in development, as well as the interactions of these processes with physiological and environmental factors, entails a revised understanding of the evolutionary role of natural selection as the presumed unique engine of adaptation. Not only the kind and number of theory elements have increased, but their inclusion also modifies the formal structure of evolutionary theory and expands its explanatory capacity. In this talk, we explore some of these new explanatory possibilities, and examine which tenets of the classical framework require revision or replacement.

11.10 – 12.25 Contributed Session C1: "Living fossils", myth or reality?

ROOM A

Chairs: Didier Casane and Patrick Laurenti

C1-01 Sharks, stems, and shedding the living fossil tag

Coates, Michael (The University of Chicago, IL, USA); **Criswell, Katharine** (The University of Chicago, IL, USA)

So-called living fossils carry phylogenetic baggage, including hypotheses about persistent and/or invariant conditions, often of interest because such features are thought to be primitive. The status of Latimeria as a living fossil has changed in tandem with related phylogenies: formerly a living tetrapod ancestor, now it tends to be compared to Cretaceous coelacanths; where once its fins were exceptional, now its genes are living fossil drivers of tetrapod evolution. Similar issues colour our interpretation of the Chondrichthyes: long used as a substantial cache of living fossils, and most recently yielding the chimaeroid Callorhinchus, apparently the slowest evolving vertebrate known. However, phylogenies framing chimaeroids and chondrichthyans are poorly resolved, and although the monophyly of these groups is undoubted, relationships of living clades to fossil sharks and

Palaeozoic jawed vertebrates in general are changing. It is increasingly apparent that conditions in the last common ancestor of crown-group gnathostomes were somewhat osteichthyan-like. More precisely, it appears that early osteichthyans include conditions that are decidedly more primitive than those in early or modern chondrichthyans. Here, we show that new phylogenetic hypotheses are supported by new discoveries of stem-lineage sharks, with direct bearing on estimates of primitive conditions for jaws, dentitions, scales, branchial arches, and other key components of gnathostome body plans. Significantly, the evolution of these systems appears to be noisier, i.e., more homoplasic, than previously thought. As for chimaeroids: they might be slow, but there is, as yet, no reason to push the origin of this clade close to the base of the chondrichthyan tree and thus close the base of modern jawed vertebrates as a whole.

C1-02 Skeletogenesis in cartilaginous fish

Enault, Sebastien (ISEM - Université Montpellier 2, FRA); Da Silva, Willian (ISEM - Université Montpellier 2, FRA); Venteo, Stephanie (INM- Université Montpellier 2, FRA); **Debiais-Thibaud, Mélanie** (ISEM Université Montpellier 2, FRA)

Cartilaginous vertebrates (chondrichthyans) include sharks, rays, skates and holocephalans, and are the sister group to osteichtyans (bony vertebrates). Most usual model organisms belong to osteichthyans but currently, more and more efforts are made to include chondrichthyan species in comparative developmental biology studies. Recent paleontological discoveries have strongly questioned the paradigm in which sharks would be a proxy for the vertebrate ancestral condition, in particular their lack of a mineralized skeleton. We still lack sets of developmental data that would help identify chondrichthyan derived characters and vertebrate ancestral characters. Here we will present our work on the description of skeletal morphogenesis in the small-spotted catshark Scyliorhinus canicula. Histological staining was used to describe different developmental units in the skeleton that display various states of calcification. We focused on comparing jaw, fin and vertebrae development. Further descriptions included the expression patterns of several members of the collagen gene family during the morphogenesis of these structures. Together these results are compared to known processes in bony vertebrate and placed in the context of early vertebrate evolution.

C1-03 *Limulus polyphemus* is not quite a "living fossil", especially from a developmental point of view

Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

Limulus polyphemus, or better all four modern species of horseshoe "crabs" (xiphosurans, euchelicerates), are often cited as an example of a "living fossil". Their morphology is thought to have evolved only little since about 450 million years. The four extant species represent a paragon of a relic group, and the general habitus is reminiscent of that of "typical" Palaeozoic arthropods, the trilobites. Also their life habit as being aquatic but breeding on the shore has often been interpreted as a kind of archaic step towards terrestrial life. Yet, I will demonstrate that the concept of living fossils (which itself seems questionable) cannot be applied to *Limulus polyphemus* (and its three close relatives). This is especially true from the developmental point of view. I present here data from fossil and extant developmental stages of different species of horseshoe "crabs". By using up-to-date imaging methods such as polarisation or fluorescence photography, stereo imaging, and micro computed tomography (microCT) it is possible to make even smallest details visible in situ. Compared to now extinct species of horseshoe "crabs", the ontogeny of the modern forms appears to be far derived. Hatchlings are at a far developed state; no pronounced larval stages are to be found. The so-called trilobite larva is in fact much further developed and much more similar to the adult than the hatchlings of the now extinct species. The modern horseshoe "crabs" represent only the peak of a once large diversity, but have significantly derived from the ancestral developmental pattern and hence should be seen as highly specialised forms instead of as "living fossils".

C1-04 Cockroach-like insects: Successful since 300 million years and therefore "living fossils"?

Hörnig, Marie K. (Ernst-Moritz-Arndt-University of Greifswald, GER); Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Joachim T. (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

Cockroaches are very persistent insects and are well adapted to numerous kinds of environment. Cockroaches are the most prominent representatives of Dictyoptera. Dictyopteran cockroach-like insects, or roachoids, were already abundant in the fauna of coal-swamp forests 300 million years ago. At first appearance, these seem quite similar to modern representatives of Blattodea (cockroaches) in general morphology and hence cockroaches might be addressed to as ‚living fossils'. But since then especially the modes of reproduction and, therefore, the morphology of the hatchlings, as well as the post-embryonic developmental pattern of dictyopterans evolved into a dramatically different direction. Before the Cretaceous (about 115 mya), representatives of Dictyoptera (s. l.) presumably laid single eggs, as indicated by a strongly elongated ovipositor. Based on numerous fossil specimens, nymphs of these early dictyopterans are rather small but far developed. First direct evidences for dictyopteran oothecae (several eggs

deposited together in a robust case; apomorphy for Dictyoptera s. str.) are from the Cretaceous, representing a first form of brood-care behaviour. Further evolution of several different reproductive modes, including a wide range of brood-care behaviour, led to quite different hatchling morphologies. Hatchlings of several species of Blattodea are blind, weakly sclerotised and therefore depend on shelter and feeding by their parents. In some species the nymphs exhibit special forms of mouthparts to cling on an adult or feed on secretions from the mother. Also more than 100 mya the origin of mantids and termites from cockroach-like ancestor represents a further deviation from the presumed conserved cockroach habitus and ontogeny. Hence modern representatives of Dictyoptera represent highly derived and adapted forms, not "living fossils".

C1-05 Segmentation in brachiopod larvae? The expression of common segment patterning genes during development of larval lobes

Vellutini, Bruno (University of Bergen, NOR); Hejnol, Andreas (University of Bergen, NOR)

Brachiopods are sessile bivalved marine invertebrates with an extensive fossil record dating back to the Cambrian. Most abundant and diverse during the Palaeozoic era, these filter-feeding animals are closely related to annelids, molluscs, and nemerteans. Development is indirect and embryos form a characteristic larva with lobes disposed along the anterior-posterior axis of the larval body. This segmented appearance has long intrigued biologists and raised questions about the homology between brachiopod larval lobes and annelid segments. In order to address this question and provide further insights into the evolution of brachiopod larval forms, we have studied the expression of genes commonly involved in the patterning of arthropod and annelid segments. We cloned Engrailed, Wnt genes, and components of the Hedgehog pathway and performed in situ hybridization in brachiopod larvae with differing morphology, the trilobed larva of *Terebratalia transversa* and the bilobed larva of *Novocrania anomala* . We observed a close spatiotemporal correlation between engrailed expression and the development of the apical/mantle lobe boundary in both larval forms. In *T. transversa*, but not in *N. anomala* , wnt1 transcripts formed an ectodermal stripe adjacent to the engrailed domain, a pattern also found in the parasegment and segment boundaries of arthropods and annelids, respectively. Other Wnt genes were expressed in a lobe specific manner suggesting a role in the patterning of larval lobes. However, this was not the case for hedgehog transcripts which were restricted to the endoderm. Our results suggest that the expression of engrailed and some Wnt genes are conserved between different brachiopod larvae and they might be involved in lobe patterning. However, there was remarkable variability of gene expression patterns at the anterior boundary and differences to typical segment patterning. Thus, we argue that there is little support for the homology between annelid segments and brachiopod larval lobes and that similarities likely originated by gene co-option during the evolution of brachiopod larval forms.

11.10 – 12.25 Contributed Session C2: Developmental basis of quantitative variation

ROOM B

Chair: Mihaela Pavlicev

C2-01 How do you shave a baby? *Cis*-regulatory region occupancy as the basis for morphological evolution

Preger-Ben Noon, Ella (Janelia Farm Research Campus, Ashburn, VA, USA); Stern, David (Janelia Farm Research Campus, Ashburn, VA, USA)

Morphology evolves by altering the timing, pattern and levels of gene expression during embryonic development. Such alterations largely result from specific DNA changes in the *cis*-regulatory regions of genes. As *cis*-regulatory regions accumulate mutations, this modifies their transcription factor binding properties and hence their transcriptional outputs. Despite extensive research in the field, it is nearly impossible to predict which transcription factors bind to a *cis*-regulatory region solely from DNA sequence. Therefore, even in cases where the causal mutations are known, how the transcriptional machinery interprets these sequence changes to produce a new phenotype is a mystery. Nor do we know the direction of the change: does evolution act through the loss of activation, gain of repression or a combination of the two? Here we address these problems by deciphering the mechanistic details underlying the regulation of evolved *cis*-regulatory sequences in the Drosophila *shavenbaby* gene. *Shavenbaby (svb)* encodes a transcription factor that controls the development of cuticular hair-like projections called trichomes. Trichome patterns have repeatedly evolved in larvae of the genus Drosophila via changes in *svb* regulatory regions. Seven enhancer modules located in the *cis*-regulatory region of *svb* together recapitulate the entire *svb* embryonic expression pattern. Multiple nucleotide changes in five of these enhancers have led to the evolutionary loss of dorsal and lateral trichomes in *D. sechellia*, a sibling species to *D. melanogaster*. Surprisingly, most of these nucleotide changes do not occur in any known transcription factor binding motifs. We combined genomic, biochemical, and genetic approaches to identify the trans-regulators that bind differentially to nucleotide sites that have evolved between *D. melanogaster* and *D. sechellia*. Our results will lead to a better mechanistic understanding of how evolved *cis*-regulatory changes are read out by the transcriptional machinery to produce a morphological change.

C2-02 Tissue sensitivity to Hox protein levels underlies the adaptive leg morphology of water striders

Refki, Peter (Institute of Functional Genomics (IGFL); UCBL, Lyon, FRA); Armisen, David (IGFL-ENS, Lyon, FRA); Crumière, Antonin (IGFL-ENS, Lyon, FRA); Viala, Séverine (IGFL-ENS, Lyon, FRA); Khila, Abderrahman (IGFL-ENS, Lyon, FRA)

The evolution of adaptive morphological traits is often associated with changes in pre-existing structures. However, the genetic mechanisms underlying these adaptive changes are poorly understood. In water striders, a group of hemipteran insects, a dramatic growth of thoracic appendages — such that T2-legs are longer than T3-legs — is associated with the specialization to locomotion on water. This excessive leg growth facilitated the exploitation of open aquatic habitats, a restricted niche for their terrestrial relatives; and hence opens a new array of opportunities. Water striders have evolved a derived mode of locomotion, which distinguishes them from ground insects, through rowing on water. They move their mid-legs in simultaneous sweeping strokes for propulsion, and move their hind-legs in fine movements for orientation. Using RNAi knockdown, gene expression analyses, and transcriptomics, we demonstrate that the emergence of rowing as a novel mode of locomotion to invade open water surface is associated with the evolution of tissue sensitivity to the dose of the Hox protein Ultrabithorax (Ubx). In the water strider *Limnoporus dissortis*, Ubx is expressed in a six to seven fold higher dose in T3-legs than in T2-legs. Ubx RNAi knockdown resulted in shorter T2-leg length that was further aggravated with increased depletion of Ubx. Dissimilarly, the knockdown phenotype on T3-legs revealed an opposite dose dependent activating-repressing effect. Ubx RNAi resulted in a longer T3-leg phenotype that was partially rescued with increased depletion of Ubx. Basal non-rowing semi-aquatic species lack this opposite dose dependence of Ubx. In addition, we demonstrate that the pattern of expression of the canonical developmental genes *distal-less (dll), hedgehog (hh), decapentaplegic (dpp), wingless (wg), epidermal growth factor receptor (egfr), dachshund (dac), homothorax (hth),* and *extradenticle (exd)* does not change in *Ubx* knockdown embryos. This indicates that the dose dependence of Ubx has no discernable effect on the spatial expression of essential leg patterning genes. On the other hand, comparative transcriptomic analyses revealed novel candidates under the control of Ubx. Our findings suggest that adaptive traits can evolve through the sensitivity of tissues to the levels of a Hox protein, due to novel interactions with downstream target genes, and independently from pattern formation during early embryogenesis.

C2-03 Developmental mechanisms underlying natural variation in organ size

Ramaekers, Ariane (VIB-KU Leuven, BEL); Weinberger, Simon (VIB-KULeuven, BEL); Buchner, Erich (University of Würzburg, GER); Wolf, Reinhard (University of Würzburg, GER); Hassan, Bassem A. (VIB-KU Leuven, BEL)

Developmental processes are highly complex, interconnected systems which translate genotypes into phenotypes, albeit in a non-linear fashion. Our main interest is to characterize the complex links between variation in developmental processes and morphology. As a model, we study the variation of the number of facets of the compound eye between and within Drosophila species. By comparing eye development between populations characterized by different numbers of eye facets - or ommatidia , we find that:
(i) The tempo and mode of eye development are largely conserved, even between relatively distant species.
(ii) The difference in facet number is associated with variation in the segregation of the head primordium into two distinct fields. This leads to the formation of a larger eye field at the expense of the antennal field. This allows the specification of a larger number of facet progenitor cells which in turn, results in more facets in the adult eye.
(iii) Similar mechanisms of developmental variation seem to underlie facet number variation in two cases we analyzed in detail: one between and one within species. This finding suggests that the mechanism we identified could constitute a favored route to eye size change in flies. Evolving sensory modalities can potentially lead to adaptation to new ecological niches. To test the functional consequences of facet number variation, we compared the visual acuity (spatial resolution) of "large" and "small" eye species using a behavioral assay. We observed that increased facet number is correlated with better acuity. This suggests a functional- and potentially adaptative – relevance of variation of this trait.

C2-04 Investigating the genetic and developmental origins of limb bone length using mice selectively bred for increased tibia length

Marchini, Marta (University of Calgary, AB, CAN); Krueger, Carsten B. (University of Calgary, AB, CAN); Sparrow, Leah M. (University of Calgary, AB, CAN); Dowhanik, Alexandra S. (University of Calgary, AB, CAN); Cosman, Miranda N. (University of Calgary, AB, CAN); Rolian, Campbell (University of Calgary, AB, CAN)

This study provides insights into the genetic and developmental origins of phenotypic variation in limb bone length using a mouse population under intensive selection on tibia length. Limb bone length is a complex quantitative trait, which involves the expression and interaction

of many genes, gene pathways and the environment. The genetic mechanisms involved in outgrowth and patterning of the vertebrate limb during embryogenesis are relatively well documented. However, how these mechanisms contribute to continuous variation in bone length remains unknown. Since 2010, our research group has selectively bred mice for increased relative tibia length. Using artificial selection in a controlled environment, and comparing selectively bred mice with unselected controls, enables the study of the developmental and genetic processes involved in producing phenotypic differences in tibia length between individuals. Specifically, it allows us to identify where, when and how the specialized cartilage cells (chondrocytes) of the growth plates in the tibia have changed, to contribute in the variation in long bone length. The growth plate is composed of distinct cellular zones reflecting different stages in chondrocytes's life cycle: resting, proliferation and hypertrophy. Preliminary histological data on the tibial growth plates suggest that 14 days old pups present differences in the size of the hypertrophic zone between selected and control mice, including the number and size of hypertrophic chondrocytes. In contrast, the rates of cell proliferation do not appear to be significantly different between the two populations. Here, using next generation RNA sequencing, we test the hypothesis that bone elongation under selection is mediated primarily through changes in the molecular regulation of hypertrophy in the tibial growth plates. RNA-seq permits a precise quantification of gene expression and the study of processes involved in bone formation between mice selectively bred for increases in limb bone length and a control group. Our results indicate that in the chondrocytes of long-limbed mice, genes such as frizzled-related protein involved in the Wnt/Beta-Catenin pathway, epiphycan (proteoglycan of hypertrophic cartilage matrix) and tenomodulin are up-regulated. The RNA-seq results also show that osteoblast and osteoclast related genes also play a significant role in mediating limb bone length. For instance in selected mice, up-regulation of beta2-microglobulin and down-regulation of osteocalcin produced by the osteoblasts seem to be involved in this mechanism, as well as the up-regulation of cytokine and chemokine expressed by hematopoietic cells. These differential expression data suggest that bone turnover might have an important role in bone elongation.

C2-05 Pattern modulation produces a highly regular grid of defensive hairs in the spiny mouse (*Acomys dimidiatus*)

Tzika, Athanasia (University of Geneva, CHE); Montandon, Sophie (University of Geneva, CHE); Manukyan, Liana (University of Geneva, CHE); Milinkovitch, Michel (University of Geneva, CHE)

The mammalian skin and its appendages exhibit a remarkable variety of phenotypes, with interspecies studies now required to uncover the developmental mechanisms generating this diversity. We investigated the development of spines in *Acomys dimidiatus*, a lineage that diverged 25 million years ago from that of the laboratory mouse. We show that the spine-forming region of the *Acomys* pelage organizes as a highly regular grid of follicle triplets generated in two steps of secondary placode induction. We use whole-mount in situ hybridizations and 3D reconstructions to investigate the processes that establish this spectacular micro-pattern. Topology/geometry statistics are employed to assess the regularity of the pattern. The impact of cell migration and proliferation inhibition is analysed on *ex vivo* skin cultures. Exome-wide gene expression comparative analyses in *Acomys* and the laboratory mouse revealed the amplification of signaling in three specific genes likely to contribute to the large *Acomys* placodes and highly derived spine-like hair morphology.

11.10 – 12.25 Contributed Session C3: Uncovering the genomic bases of phenotypic change in the NGS era I

ROOM C1

Chairs: Manuel Irimia and Juan Pascual

C3-01 Two novel, complementary next generation sequencing approaches to reveal the dorso-ventral gene regulatory network of *Tribolium castaneum*

Stappert, Dominik (University of Cologne, GER); Frey, Nadine (University of Cologne, GER); Roth, Siegfried (University of Cologne, GER)

One of the best-understood gene regulatory networks (GRN) is governing patterning along the dorso-ventral (DV) axis of the fruit fly Drosophila. This network is dominated by Toll signaling. Interestingly, Toll signaling has a broadly conserved function in innate immunity, whereas its function in DV patterning seems to be a novelty: In most animals not Toll, but BMP signaling is essential for DV patterning while in Drosophila BMP signaling has a limited role downstream of Toll signaling. From an evolutionary perspective, Toll signaling must have acquired functions in DV patterning during insect evolution, culminating in the molecular phenotype observed in Drosophila. Thus, studying Toll signaling in different insects promises a detailed understanding of GRN evolution. The Drosophila DV GRN serves as gold standard to which other insect DV GRN will be compared. Former insights into insect DV patterning were mainly achieved by candidate gene approaches. One current challenge is to overcome this approach by identifying genes not only by virtue of their homology to Drosophila, but in an unbiased manner. To that end we present a combined wet-lab and bioinformatics approach: the establishment and use of chromatin immune precipitation followed by sequencing (ChIP-seq) in combination with RNA interference followed by comprehensive differential expression

analysis (RNA-seq) for the elucidation of DV GRN components in the beetle *Tribolium castaneum*. ChIP-seq for a transcription factor was not performed before in a non-model organism. In combination with RNA-seq it allowed the identification of 40 bona fide *Tribolium* DV patterning genes, some of which are unknown from the Drosophila DV GRN. Analysis of these 40 bona fide genes will enhance our knowledge on the function of the *Tribolium castaneum* DV patterning system. Further, our results offer a strategy of how to approach GRNs in non-model species in an unbiased, global manner enabling molecularly detailed evolutionary comparisons.

C3-02 Convergent evolution of proteins with repetitive, low complexity domains in biomineralizing taxa

McDougall, Carmel (University of Queensland, Brisbane, AUS); Woodcroft, Ben (University of Queensland, Brisbane, AUS); Degnan, Bernard (University of Queensland, Brisbane, AUS)

Proteins that are comprised of highly repetitive and modular sequences have increasingly been identified from the matrices of calcium-based biomineralised structures, and have been likened to spider silks that are renowned for their exceptional strength and elasticity. The common repetitive motifs relate to the secondary structure of the protein and are the ultimate key for the development and physical properties of these extracellular structures. Given these common sequence characteristics, we developed a bioinformatics-based method to identify candidate biomineralisation genes from large-scale sequence databases. We have applied this method to several datasets to identify novel silk-like genes that appear to have evolved convergently in numerous animal lineages. Genes identified by this technique are expressed in cells responsible for the development of the sea urchin larval spicule, and in the molluscan mantle that is responsible for shell secretion, consistent with their role in biomineralisation in these bilaterians. Given the diversity of structures with which these sequences are involved, the genetic distance of the organisms they are found in, and their rapid evolutionary rates, we propose that biomineralisation proteins with highly repetitive, modular sequences evolved multiple times independently in different metazoan lineages.

C3-03 Cnidarian microRNAs frequently regulate their targets by cleavage

Praher, Daniela (University of Vienna, AUT); Moran, Yehu (The Hebrew University of Jerusalem, ISR); Fredman, David (University of Vienna, AUT); Li, Xin (University of Massachusetts, Worcester, MA, USA); Wee, Liang-Meng (University of Massachusetts, Worcester, MA, USA); Rentzsch, Fabian (University of Bergen, NOR); Zamore, Philip (University of Massachusetts, Worcester, MA, USA); Seitz, Hervé (CNRS, Montpellier, FRA); Technau, Ulrich (University of Vienna, AUT)

The evolution of complexity is a long-standing issue in evolutionary biology. In recent years, the phylum Cnidaria (sea anemones, hydras, corals and jellyfish) has gained broad interest in comparative studies due to its phylogenetic position as the likely sister group to Bilateria. In this context, the sea anemone *Nematostella vectensis* has emerged as the leading cnidarian model system due to several advantages like the availability of a sequenced genome and molecular manipulation techniques. *Nematostella* has a surprisingly similar gene repertoire to bilaterians including gene structure and *cis*-regulation of transcription. Despite these similarities, *Nematostella* has a much simpler morphology than most bilaterians. Since the cause for the difference might lie in the post-transcriptional regulation of gene expression, we investigate microRNAs (miRNAs) in *Nematostella*. Although much knowledge about the physiological and developmental importance of miRNAs has been gained in bilaterian and plant model organisms, their evolution remains largely elusive. In Bilateria, miRNAs regulate most of their target transcripts by binding to them via a very restricted stretch of 6-7 nucleotides, the seed sequence. This promotes translational inhibition and transcript decay. In contrast, most plant miRNAs show a near perfect complementarity to their targets leading to cleavage. We sequenced small RNAs from different developmental stages and learned that they are spatiotemporally regulated like in Bilateria and plants. Interestingly, *Nematostella* miRNAs show an extensive complementarity to their targets, expanding beyond the seed sequence, a situation reminiscent of small interfering RNAs (siRNAs) and plant miRNAs but highly uncommon for bilaterian miRNAs. We showed by targeted as well as genome wide approaches that this high complementarity leads in many cases to miRNA-directed cleavage of target mRNAs in a similar manner to plant miRNAs and siRNAs. This miRNA-directed cleavage mechanism may be common to Cnidaria since we find it in another, distantly related species. The mechanistic similarities between plant and cnidarian miRNAs as well as siRNAs, which are considered to be ancestral small RNAs, lead us to propose that extensive complementarity and cleavage might represent the ancestral state of small RNA mediated target regulation. Our data indicate that miRNA-mediated regulation functions very differently in Cnidaria and Bilateria and that seed-based target regulation may be an innovation of bilaterian miRNAs. Thus we hypothesize that this contributes to the differences in morphological complexity between these two animal groups.

C3-04 Trancriptome profiling of a key morphological innovation: The propelling fan of the water walking bug, *Rhagovelia obesa*

Santos, Emilia (Institute of Functional Genomics (IGFL), Lyon, FRA); Khila, Abderrahman (IGFL, Lyon, FRA)

The invasion of new habitats often requires the evolution of key novel traits that will facilitate the exploitation of the new environment. The origin of novel phenotypic characters is therefore a key component in organismal diversification, yet the mechanisms and selective forces underlying the emergence of such evolutionary novelties are largely unknown. In Rhagovelia sp. (genus of water-walking insects) the evolution of a highly elaborate swimming fan on the tarsus of the propelling mid-legs increases water resistance against leg movements, thereby increasing their propelling function. This novel adaptive trait allowed the Rhagovelia group to conquer and diversify on running water surfaces — a niche that is not accessible for most other water-walking insects. However, the genes underlying the differentiation of the fan during development are unknown. With next-generation sequencing it is now possible to access transcriptomes and gene expression at unprecedented levels. Here we first report a comparative analysis of the full transcriptomes of *Rhagovelia obesa* and *Limnoporus dissortis*, another water walking insect without propelling fans. Second, we provide a characterization gene expression levels between developing mid-legs (with fan) against forelegs and hind-legs (without fan) in *R. obesa*. We further compare gene expression profiles between the legs of *R. obesa* and those of *L. dissortis*. This approach has allowed the identification of a list of candidate genes associated with the evolution of this strikingly novel phenotype. Finally, we provide evidence that both co-option of pre-existing genes and lineage specific genes might be involved in the evolution of novel phenotypes. Studying the developmental genetic and adaptive bases of this phenotype will help understand the mechanisms through which spectacular morphological adaptations can arise under specific environmental pressures.

C3-05 Phenotypic plasticity and epigenetics in the honeybee ovary

Leask, Megan (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL); Duncan, Elizabeth (University of Otago, Dunedin, NZL)

Phenotypic plasticity is a widespread phenomenon that allows an organism to change its phenotype in response to the environment. This plastic response is evident throughout animal phyla and has likely persisted because it allows the organism to enhance its fitness. Phenotypic plasticity is therefore an important concept to explore in regards to understanding the mechanisms that underlie evolution and development.

Honeybees display a remarkable example of phenotypic plasticity. In the presence of the queen and the environmental stimulus queen mandibular pheromone (QMP) worker bee ovaries are maintained in a stable inactive state. Removal of the queen, and thus QMP, causes a shift in the physiological state of the worker ovary, with changes in gene expression resulting in the physical remodeling and activation of the worker bee ovary. In this research the honeybee worker ovary has been used as a model of phenotypic plasticity in order to determine the role epigenetic mechanisms play in establishing or maintaining phenotypic plasticity.

Gene expression analyses have identified that genes with roles in chromatin remodelling are differentially expressed between queen, active, and inactive worker ovary. Chromatin immunoprecipitation has been applied in conjunction with high-throughput sequencing to identify genome-wide epigenetic changes that occur during the process of ovary activation. In addition functional testing through the inhibition of particular histone modifications has revealed a key role for epigenetic modifications in establishing phenotypic plasticity.

11.10 – 12.25 Contributed Session C4: Extended Evolutionary Synthesis & Quo vadis EvoDevo?

ROOM C2

Chair: Werner Callebaut

C4-01 The origination of novelty: Qualitative changes from quantitative variation

Peterson, Tim (University of Vienna, AUT); Müller, Gerd B. (University of Vienna, AUT)

An important aspect of EvoDevo is not only how development evolves, but also how development itself can impact evolution. Due to the procedurally generated nature of development, qualitative discontinuous changes can occur as side effects of crossing thresholds in quantitative variation. The resultant change is dependent on the developmental properties of the system, and may later become refined by natural selection. To understand how these discontinuous changes occur, an understanding of both the development of an organism and the potential of tissue interactions are necessary. We demonstrate a technique for analyzing the biomechanics involved during the development of two species of Cichlidae to understand how these forces impact the origination of a novel joint in the pharyngeal jaw apparatus caused by cyclic compressive force. MicroCT scans are used to create 3D finite element models of varying stages in ontogeny to determine how the biomechanical forces change throughout development. By individually adjusting the various model parameters to match ancestral conditions, we can test the extent to which the biomechanics of the extant phenotypes are influenced by three derived traits of the pharyngeal jaw apparatus: a new muscle sling, a fused lower pharyngeal jaw, and a decoupling

of the upper and lower pharyngeal jaws. We show that increased pressure between the neurocranium and the upper pharyngeal jaws is caused primarily by the decoupling between epibranchial 4 and the tooth-bearing pharyngobrancials. The new muscle sling increases bite force by altering force vector orientation, also contributing to stress on the neurocranium. The fused lower pharyngeal jaws are used for force concentration on a prey item, but are not a factor in joint origination. This demonstrates the generative potential of procedurally generated development in creating novel structures, and how EvoDevo is more than the evolution of development.

C4-02 Adaptive dynamics modelling with evolving epigenetic switches

Van Dooren, Tom J. M. (Institute of Ecology and Environmental Sciences Paris, FRA)

A long-term perspective on fitness and evolutionary dynamics is essential to understand whether an epigenetic architecture is adaptive. I propose to use invasion fitness of mutants and adaptive dynamics approximations with explicit genotype-phenotype maps to investigate this. However, when many traits are evolving in these approximations, we need to carry out large-scale explorations of the structure of high-dimensional trait spaces similar to what is required for the theoretical study of genotype networks. A specific classic model in evolutionary ecology, with soft selection in two types of patches, is extended with developmental plasticity and epigenetic resetting in gametes. Ten traits are evolving in this model. Classic results for the occurrence of genotype polymorphisms are recovered, but this time for epigenotypes. The analysis of adaptive dynamics approximations indicates that there are extensive neutral networks for some traits, and that often the dynamics of traits controlling epigenetic switches will converge to the boundaries of trait space. At some of these boundaries, evolutionary dynamics can be very intricate, with strong effects on fitness of which epigenetic state an individual is in when its genotype mutates. The approach proposed incorporates many elements that are ranked high on the list of concepts or mechanisms to be included in the new Extended Evolutionary Synthesis. The results on one of the simplest models of evolutionary ecology suggest that adaptive storytelling will become an exclusive feature of the longer versions of theoretical papers, due to its intricacies.

C4-03 The timing of development

Nicoglou, Antonine (Institut d'Histoire et de Philosophie des Sciences et des Techniques, Paris, FRA)

The issue of whether and how a gathering of developmental and evolutionary explanations should be achieved raises difficulties. In a certain way, one could consider that "problems concerned with the orderly development of the individual are unrelated to those of the evolution of organisms through time" (Wallace 1986). Since development can be depicted as the trajectory of an individual from the zygote stage to the adult stage, in a process in time, at least its timescale appears clearly decoupled from the evolutionary timescales (Hall & Olson 2006). Furthermore, the developmental process may include various processes at distinct time and space scales (molecular, cellular, etc.), which can be further analyzed on its own. I suggest that by focusing on the *character* instead of the developmental stage (de Beer 1940), developmental biology has lost the temporal dimension of its process. I argue that a way to reassess the importance of time in developmental process (in order maybe to achieve afterwards a gathering of development and evolution) is to addresses the specifics of the developmental timing, its specificities and its relation to other time scales. This would, first, offer a clarification of the separation between evolutionary and developmental timescales and then show how a developmental theory might integrate the various processes at distinct space and time scales that I identify.

C4-04 How a better understanding of developmental mechanisms would transform the bases of evolutionary biology

Salazar-Ciudad, Isaac (Institute of Biotechnology, Helsinki, FIN)

At least since the early 1980s (from Alberch and others" work) it has been clear that phenotypic variation is not possible in all directions and that which phenotypic variation is possible in an organ depends on developmental dynamics. Our understanding of the mechanisms of development has improved substantially since the early 1980s. Yet, evolutionary theory as a whole has only marginally benefited from these improvements. Central to this quest is a better understanding of the relationship between genotype and phenotype and the capacity to predict the type of phenotypic variation possible in an organ or species. I will present how this is already possible in some organs and how work this provides general views about the how development affects morphological evolution. However, any insights about morphological evolution won from understanding development are limited by our capacity to understand how development itself evolves. There are some reasons to expect, however, that there are a limited number of ways in which development can work to produce complex phenotypes. If so, the evolution of development can be understood by looking at how these replace each other over evolutionary time. These replacements depend on how likely is a development mechanisms to arise by mutation and how likely it is to produce adaptive variation in a given environment. We will show models where this happens and would explain how these can be used to make address questions

that are possible, or "no-questions", from evo-devo-free evolutionary theory. This is, for example, which types of development, or which changes in development, are most often associated with morphological novelty, how development will change when specific types of selective pressures act on morphology, etc.

C4-05 Computing the concept of evolvability

Nuño de la Rosa, Laura (The KLI Institute, Klosterneuburg, AUT)

Although EvoDevo is a well-established discipline, there is no general agreement on the impact of the introduction of development into the general structure of evolutionary theory. Instead of tackling this issue from a theoretical perspective about how development should be integrated in evolutionary theory, this paper takes a computational approach to the history and philosophy of science with the aim of exploring in a quantitative manner how development has been actually integrated in evolutionary biology. In accordance with the claim that philosophy of science should shift the emphasis from theories to problems or epistemic goals, I will focus on the concept of *evolvability*, which is usually taken to be a cornestone of EvoDevo and, more generally, of an Extended Evolutionary Synthesis (EES). In particular, I will use some of the quantitative techniques developed in science studies to map different aspects of the structure and dynamics of scientific fields. First, I will look at the recent history of EvoDevo by mapping emerging trends and abrupt changes in the research front of evolvability. Second, I will attempt to reconstruct the conceptual structure of EvoDevo departing from the notion of evolvability. I will combine co-citation and co-word analysis to identify the main resarch topics within the field of EvoDevo and how they relate to other research topics such as modularity or the genotype-phenotype map. Finally, I will analyze the role played by the notion of evolvability in the integration of development into evolutionary theory. On the one hand, I will analyse in a comparative framework the evolution of the percentages of the research areas related to the papers on evolvability. On the other hand, I will apply journal co-citation analysis to explore in more detail how evolvability has influenced the relationship between developmental biology and two key disciplines in the constitution of an EES, namely computational evolutionary biology and population genetics.

11.10 – 12.25 Contributed Session C5: Developmental mechanisms underlying evolutionary change I

ROOM D

Chair: Constanze Bickelmann

C5-01 Hox expression in salamanders: Preaxial polarity revisited

Bickelmann, Constanze (Museum für Naturkunde, Berlin, GER); Schneider, Igor (Instituto de Ciencias Biologicas, Belem, BRA); Fröbisch, Nadia B. (Museum für Naturkunde, Berlin, GER)

Digit development is highly conserved among tetrapods and is characterized by the formation of the digital arch proceeding in preaxial direction. Salamanders are the only exception, showing a reversed postaxial direction in digit development. This and other peculiarities make salamanders an ideal non-model organism in which to study limb development; surprisingly, molecular data is scarce and largely limited to regenerating tissues. Elucidating the mechanisms and molecular trajectories of digit development in taxa showing conserved patterns as well as in taxa displaying variant pathways is required to gain a coherent picture of the evolution and development of the tetrapod limb in general. Here, we report a comprehensive re-analysis of *HoxA11* expression in the developing limbs of the axolotl *Ambystoma mexicanum*. *HoxA11* is known to play a central role in the specification of the zeugopodial elements in model organisms. A previous study reported a different autopodial expression of *HoxA11* in salamanders, in which digits 1 and 2 formed in the absence of *HoxA11* expression, while expression occurred in the central autopod as well as in developing postaxial digits 3 and 4. Based on this, it was suggested that the postaxial digits of salamanders might represent re-evolved de novo structures derived from zeugopodial precursors after an evolutionary phase of digit reduction in salamanders. In this study, we could confirm expression of *HoxA11* in digits 3 and 4 in axolotl autopods. The scenario of postaxial de novo digits, however, is not supported by data from the fossil record of the urodelan lineage. In order to better understand the evolution of the unique expression of *HoxA11*, we further investigate expression of additional Hox genes that are known to play a central role in digit formation and identity; preliminary results from ongoing analysis of *HoxD13* expression are also presented.

C5-02 An embryological perspective on the lung structure of early amniotes

Lambertz, Markus (University of Bonn, GER); Grommes, Kristina (University ofBonn, GER); Kohlsdorf, Tiana (Universidade de São Paulo, Ribeirão Preto, BRA); Perry, Steven F. (University of Bonn, GER)

An effective respiratory system for air breathing was one of the key elements in the course of vertebrate terrestrialization, i.e., the rise of

amniotes. The traditional textbook view on this crucial evolutionary transformation is that the early amniotes exhibited lungs similar to those of extant amphibians: sac-like, single-chambered organs. Although mammals, turtles, crocodiles and birds have a rather complex, multichambered/branched pulmonary anatomy, this scenario seems to be confounded by the fact that the majority of lepidosaurs indeed have single-chambered lungs. We show that lepidosaurs develop such pulmonary complexity during early ontogeny as well, which, however, eventually becomes secondarily reduced and obscured. Our results consequently will require a rethinking of the hypothesis about the early evolution of the respiratory apparatus among amniotes. Terrestrialization apparently was accompanied from the very beginning not only by aspiration breathing but also by complex lungs, and the single-chambered lungs of lepidosaurs represent a derived condition. We provide an evolutionary scenario for the evolution of lungs among amniotes that is based on extensive comparative anatomical and embryological data and acknowledges the fossil record and the biophysical constraints that act on pulmonary structure.

C5-03 The intricate relationship between selection and developmental constraints: the evolution of the Drosophila sex comb length

Malagon, Juan (University of Toronto, CAN)

In spite of the diversity of possible biological forms observed in nature, a limited range of morphospace is frequently occupied for a given trait. Several mechanisms have been proposed to explain this bias including selection, drift and developmental constraints. Many studies have experimentally shown how selection and drift can produce this phenotypic bias. However, most of the proposed examples of developmental constraints lack experimental evidence due to a poor understanding of the underlying mechanisms. We studied the male sex comb, a group of modified bristles used in courtship that shows spectacular morphological diversity among Drosophila species. In many Drosophila species, including *Drosophila melanogaster,* the sex comb rotates 90° to a vertical position during development. Here we analyze the effect of changing sex comb length in *D. melanogaster* and provide evidence for an internal constraint in achieving long vertical combs. To do so, we integrate three different approaches, genetic manipulations through artificial selection and mutations, cellular-developmental analysis based on live imaging, and comparative morphology of closely related species. We find that artificial selection is able to increase sex comb tooth number. However, rather than producing the relatively straight sex comb shape observed in other Drosophila species, the sex comb bends due a mechanical restriction imposed by a neighboring row of bristles. Our result shows an example of a conflict between selection and development and different ways in which this problem is solved among Drosophila species. In addition, this work demonstrates how simple physical principles can play a role in biasing the direction of evolution.

C5-04 A burst of microRNA innovation in the early evolution of butterflies and moths

Quah, Shan (University of Oxford, GBR); Holland, Peter (University of Oxford, GBR)

MicroRNAs (miRNAs) are known to be involved in the post-transcriptional regulation of gene expression during development. Studying their evolution may offer insights into the evolution of development, and hence shed light on morphological innovation. We sought to clarify the distribution of miRNAs within the Lepidoptera (butterflies and moths). This is an ideal group in which to investigate miRNA evolution due to a plethora of genomic resources and miRNA sequencing studies. We sequenced small RNA libraries in a basally divergent lepidopteran, *Cameraria ohridella* (Horse-chestnut Leafminer), as well as in a representative of the highly speciose nymphalid family, *Pararge aegeria* (Speckled Wood butterfly), and mapped sequences to genomes. 90 and 81 conserved miRNAs were identified in *C. ohridella* and *P. aegeria* respectively, along with a large number of species-specific miRNA genes. Mapping these data onto a phylogeny reveals a burst of miRNA innovation early in lepidopteran evolution, suggesting that miRNA acquisition accompanied the early radiation of the Lepidoptera. Novel lepidopteran miRNAs are expressed at a comparable level to more ancient sequences, which is suggestive of functional integration into transcriptional networks. One of these miRNAs may have a conserved role in the regulation of segmentation and wing patterning.

C5-05 Stem cell genes characterization in *Oscarella lobularis* (Porifera, Homoscleromorpha): The stepping stone towards understanding somatic and germ lines origin

Fierro, Laura (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); Schenkelaars, Quentin (IMBE, Marseille, FRA); Borchiellini, Carole (IMBE, Marseille, FRA); Ereskovsky, Alexander (IMBE, Marseille, FRA); Renard, Emmanuelle (IMBE, Marseille, FRA)

Stem cell research is one of the major issues of modern developmental biology but also of regenerative medicine. Here, we focus on the homoscleromorph sponge *Oscarella lobularis*. Since sponges are one of the most basal metazoan groups, they hold a key position to address the stem cells origin and the distinction between somatic and germinal cell lineages in early metazoan evolution. It is commonly accepted in sponges that archaeocytes are the poriferan stem cells and they are source of both, somatic and germ cell lineages. In addition, the choanocyte, involved in water filtration, can also give

rise — by transdifferentiation — to gametes in some sponge species. Thereby, unlike most metazoans there is no permanent distinct germ line-somatic separation. The absence of archaeocytes in *O. lobularis* suggests that its reproductive strategy is only based on the choanocyte ability to transdifferentiate. Therefore, we investigate the mechanisms underlying sexual reproduction in *O. lobularis* by studying proteins known to be conserved in other metazoans as the germline molecular machinery (Piwi, Boule, Vasa, PL10, Nanos, Bruno, etc.). These proteins are mainly involved in transcriptional repression of somatic programs and post-transcriptional regulation during gametogenesis or/and embryogenesis. Candidate genes mentioned above were searched in *O. lobularis* databases. Homologs of germ line marker genes were identified by Blast approaches combined with phylogenetic and protein domain analyses. Hence, these specific genes were conserved during metazoan evolution. Furthermore, in situ hybridization results show that most of these genes expression are located not only in choanocytes during gametogenesis but also in embryos. This suggests that germ cell specification and differentiation mechanisms are shared across metazoans. Moreover, these results give even more strength to recent hypotheses proposing a common evolutionary origin of the germ line and multipotent somatic stem cells. This work highlights the relevance of Porifera to understand the processes in stem cell and germ cell biology. Besides potential interest in cellular and medical fields, we plan to study the mechanisms allowing adaptive strategies of reproduction in response to environmental changes.

14.00 – 15.40 Symposium S5: Mechanical mechanisms of development II

ROOM A

Organizers: Naomi Nakayama and Annemiek Cornelissen
Chairs: Naomi Nakayama and Annemiek Cornelissen

S5-01 Directional mechanical signals add robustness to plant morphogenesis

Hamant, Olivier (ENS Lyon, FRA)

Changing shape is changing structure. This implies that at any given time point, a shape can be associated with a pattern of tension and compression, i.e., a pattern of mechanical stress. As shown mainly in animal single cells, mechanical cues can affect important cell processes such as cell polarity, cell fate or cell division. Plants are ideal systems to investigate how mechanical signals control development in a tissue context, for technical reasons but also as mechanics in plants mainly relies on the balance between cell wall stiffness and turgor pressure. Focusing on the shoot apical meristem, the plant stem cell niche, we found that mechanical signals control the orientation of cortical microtubules, which guide the deposition of cellulose and thus control the mechanical anisotropy of plant cell walls. This in turn supports morphogenetic events such as tissue folding that further consolidate the stress pattern. We also found that this mechanical feedback loop promotes growth heterogeneity in tissues. We propose that the maintenance of a basal level of growth heterogeneity potentiates organogenesis. Scaling up, we have analyzed the link between mechanical stress and the pattern of organ initiation at the shoot meristem. Whereas this pattern seems relatively independent from mechanical inputs during initiation, a novel and robust post-meristematic pattern was obtained when microtubules are genetically uncoupled from the cellulose deposition machinery. This provides a didactic example of the interplay between growth and patterning, and suggests a contribution of mechanical stress in the robustness of patterns. Prospects for this work are numerous and will be discussed in the talk.

S5-02 Mechanics of cell contacts during tissue morphogenesis

Lenne, Pierre-François (IBDM, Marseille, FRA)

Force transmission in tissues requires adhesion between cells. Using quantitative imaging and force measurements in vivo, we study how cell-cell contacts are organized and how subcellular tensile forces are transmitted at adhesive clusters to shape cells in tissues. We will present how local contractile forces produce local and global cell shape changes during morphogenesis of early epithelia.

S5-03 Mechanical development of veins controls shape, position and movements of plants leaves

Douady, Stéphane (Université Paris-Diderot / CNRS, FRA)

Plants started to explode when they invented stems to spread their leaves above each other. However, this is possible only with the simultaneous invention of veins, both to perfuse and drain the leaves, and prolong these flows along the stem. The evolved stem itself, constituted of wood, can be seen as secondary fusion of veins, and the lignified tissue around the veins being also responsible for the mechanical rigidity of the leaf. Here we want to show that the development of these veins is often not homogeneous and constant in time. For the embryonic leaf, it can imply the formation of folds. In turns, these folds move the developing lamina, which development is being mechanically restricted by the other leaves. This induces the effective shape of the leaf. But eventually end up flat, horizontal and the adaxial face upward. This is achieved coming out from the bud with many movements, from exaggerated unfolding to nutations. These movements can be related again to the inhomogeneous growth of the veins. They can be preserved on the mature leaf as sensitivity or circadian movements. The origin of

these movements of various and unclear phenotypic interests can thus be seen as the preservation of a mechanical consequence of archaic inhomogeneous growth.

S5-04 Mechanical basis of seashell morphogenesis

Moulton, Derek (University of Oxford, GBR); Goriely, Alain (University of Oxford, GBR); Chirat, Régis (Université Lyon 1, Villeurbanne, FRA)

Seashells have intrigued scientists and mathematicians alike for centuries. Over the past several hundred years paleontologists have amassed a huge body of observations on shell form and its evolutionary variation and diversification. However, though the patterns in shells have been well documented, the formation of those patterns has only been explained in functional terms, while the developmental mechanisms underlying shell morphogenesis have remained largely elusive. I will present work on mechanical models for shell morphogenesis that we have developed, based on generic fundamental mechanics of the growth process. In particular, we demonstrate a natural, physical basis for the nearly ubiquitous presence of shell ornamentation such as spines and ribs.

14.00 – 15.40 Symposium S6: EvoDevo of symmetry in animals and plants

ROOM B

Organizers: Sophie Nadot and Catherine Damerval
Chair: Catherine Damerval

S6-01 Do asymmetric flies fly in circles? Functional consequences of a genetically induced asymmetry on flight performance in *Drosophila melanogaster*

Debat, Vincent (UMR7502 ISyEB, Museum National d'Histoire Naturelle, Paris, FRA); Aponte, Jose David (Florida State University, Tallahassee, FL, USA); Cornette, Raphaël (UMR7502 ISyEB, Museum National d'Histoire Naturelle, Paris, FRA); Herrel, Anthony (UMR7504 FUNEVOL, Museum National d'Histoire Naturelle, Paris, FRA); Peronnet, Frédérique (UMR Biologie du Développement, UPMC, Paris, FRA)

We recently discovered that, in *Drosophila melanogaster*, transgenic deregulation of Cyclin G, a protein involved both in transcriptional regulation and the cell cycle, generates extreme fluctuating asymmetry in various traits, including the wing. Fluctuating asymmetry is the subtle, random deviation from perfect symmetry that occurs in any bilateral trait, and results from developmental noise. Our results are interesting for two reasons: (1) they suggest that Cyclin G plays an important role in the control of fluctuating asymmetry, whose genetics has been elusive for decades; (2) they provide a unique tool to experimentally generate asymmetric flies — yet otherwise phenotypically normal — allowing to investigate the effects of wing asymmetry on flight performance. In this talk I will present our first results from an experimental set up allowing to film asymmetric — and less asymmetric — transgenic flies deregulating Cyclin G, extract trajectories and quantify flight parameters (velocity, acceleration, sinuosity, etc.). Coupled with a precise quantification of wing shape and wing asymmetry, these flight analyses suggest that asymmetric flies do not necessarily fly in circles.

S6-02 Role of myosin ID in Drosophila and zebrafish left-right asymmetry

Noselli, Stephane (IBV - CNRS UMR7277, Nice, FRA)

Breaking left-right (L/R) symmetry in Bilateria embryos is a major event in body plan organization. T he establishment of L/R asymmetry is essential for handedness, directional looping of internal organs (heart, gut…) and differentiation of the heart and brain. Defects in L/R asymmetry during embryogenesis can lead to a variety of defects including congenital heart diseases, spontaneous abortion, asplenia, polysplenia, etc. In vertebrates, L/R asymmetry can be set up at different stages during embryonic development, involving distinct mechanisms including the nodal flow, ions flux and asymmetric cell migration. In order to better understand how L/R asymmetry is established in protostomes, we initiated the study of L/R asymmetry in Drosophila. Our work identified the rotation of genitalia as a suitable L/R phenotypic marker: during metamorphosis, genitalia undergo a stereotyped 360° clockwise (or dextral) rotation leading to the coiling of the spermiduct around the gut. The conserved *myosinID* gene (*myoID, akaMyo31F*) was identified as a major L/R determinant in flies, required for dextral coiling of organs including genitalia and gut. In the absence of *myoID* gene activity, flies show a *situs inversus* phenotype and organs undergo sinistral morphogenesis. We will present recent results showing the role of the HOX gene *Abdominal-B (Abd-B)* in controlling early establishment of L/R asymmetry, distinct from its function in anterior-posterior patterning. *Abd-B* mutant flies develop *symmetrically*, with *Abd-B* controlling the expression of the myoID dextral determinant as well as the activity of the opposite sinistral pathway. Therefore, *Abd-B* acts as an upstream factor controlling the transition from symmetry to asymmetry in flies. We will also present recent results showing the role of zMyoID in zebrafish L/R asymmetry. Altogether, our data provide new information on the upstream mechanisms establishing L/R asymmetry in Drosophila as well as revealing a conserved function of MyoID in vertebrates L/R asymmetry.

S6-03 Corolla monosymmetry: Evolution of a morphological novelty in the angiosperms

Zachgo, Sabine (Osnabrück University, GER)

Evolution of floral monosymmetry is thought to be a major driving force of angiosperm radiation, making flowering plants the most successful land plant group in terms of species richness. Monosymmetry evolved from a polysymmetric ancestor repeatedly in different angiosperm lineages, where it likely facilitated diversification through the interaction with insects. In the Brassicaceae, only few members develop a monosymmetric corolla with two petal pairs of unequal size, making them an ideal system to study the evolution of molecular mechanisms enhancing flower complexity. Formation of corolla monosymmetry in distantly related eudicot taxa has been shown to be controlled by *TCP* transcription factors that belong to the *CYC2* clade. *CYC2* expression analyses from *Iberis*, *Calepina* and *Teesdalia* and respective heterologous overexpression studies in Arabidopsis unraveled the importance of *CYC2* expression changes to establish corolla monosymmetry, likely by affecting cell division regulation. Flower monosymmetry evolved first in the basal angisoperms, namely in the Piperales. *TCP* genes were isolated from the genus *Aristolochias l.* as it comprises over 400 species with novel and elaborate perianth morphologies such as monosymmetric tubular flowers. Expression and overexpression studies were conducted with *TCP* genes from Aristolochia arborea, which forms a highly specialized and unique mushroom mimicry in the perianth, a morphological novelty attracting pollinators to their flowers.

S6-04 *CYCLOIDEA*-like genes and floral symmetry in Ranunculaceae

Jabbour, Florian (National Museum of Natural History, Paris, FRA); Cossard, Guillaume (University of Lausanne, CHE); Le Guilloux, Martine (CNRS, Gif-sur-Yvette, FRA); Sannier, Julie (Université Paris-Sud, Orsay, FRA); Nadot, Sophie (Université Paris-Sud, Orsay, FRA); Damerval, Catherine (CNRS, Gif-sur-Yvette, FRA)

Floral bilateral symmetry (zygomorphy) has evolved several times independently in angiosperms from radially symmetrical (actinomorphic) ancestral states. Homologs of the *Antirrhinum majus CYCLOIDEA* gene (*CYC*) have been shown to control floral symmetry in diverse groups in core eudicots. In the basal eudicot family Ranunculaceae, there is a single evolutionary transition from actinomorphy to zygomorphy in the ancestor of the tribe Delphinieae. We characterized *CYC* homologs in 18 genera of Ranunculaceae, including the four genera of Delphinieae, in a sampling that represents the floral morphological diversity of this tribe, and reconstructed the evolutionary history of this gene family in Ranunculaceae. Within each of the two *RANACYL* (Ranunculaceae *CYCLOIDEA*-like) lineages previously identified, an additional duplication possibly predating the emergence of the Delphinieae was found, resulting in up to four gene copies in zygomorphic species. Expression analyses indicate that the *RANACYL* paralogs are expressed early in floral buds and that the duration of their expression varies between species and paralog class. At most one *RANACYL* paralog was expressed during the late stages of floral development in the actinomorphic species studied whereas all paralogs from the zygomorphic species were expressed, composing a species-specific identity code for perianth organs. The contrasted asymmetric patterns of expression observed in the two zygomorphic species is discussed in relation to their distinct perianth architecture.

14.00 – 15.40 Symposium S7: The Roche Discovery Oncology Symposium: Perspectives on Wnt signaling

ROOM C1

Organizers: Wim Damen and Cornelius Eibner
Chairs: Wim Damen and Cornelius Eibner

S7-01 Cancer mutations derail Wnt signalling via conformational conversion of the scaffold protein Axin

Maurice, Madelon (Utrecht University, NLD); Anvarian, Zeinab (Utrecht University, NLD); Nojima, Hisashi (MRC National Institute for Medical Research, Mill Hill, London, GBR); Madl, Tobias (Utrecht University, NLD); Spit, Maureen (Utrecht University, NLD); van Kappel, Eline (Utrecht University, NLD); Scherpenzeel, Revina (Utrecht University, NLD); Low, Teck Y. (Utrecht University, NLD); Kuper, Ineke (Utrecht University, NLD); Jordens, Ingrid (Utrecht University, NLD); Gerlach, Jan P (Utrecht University, NLD); Heck, Albert J. R. (Utrecht University, NLD); Vincent, Jean-Paul (MRC National Institute for Medical Research, Mill Hill, London, GBR); Rüdiger, Stefan G. D. (Utrecht University, NLD)

Missense mutations in multidomain tumour suppressors are frequent in human cancer, but their clinical significance and mode of action in tumour development remain poorly understood. I will discuss our findings that cancer point mutations in the scaffold protein Axin derail Wnt signalling and promote epithelial tumourigenesis *in vivo*. We found that Axin acquires pro-tumorigenic activity through structural destabilisation and subsequent oligomerisation of its N-terminal RGS domain. Non-aggregated, natively disordered regions of Axin extend away from the oligomeric core and act as molecular tentacles that

mediate aberrant interactions with a wide range of partner proteins. Mutation of aggregation-prone regions in the RGS domain prevents oligomerisation and rescues tumour suppressor activity of the mutant protein *in vivo*. Thus, the newly gained activity rather than loss of domain function underlies the pro-tumorigenic properties of the mutant protein. We propose that conformational conversion of a scaffold protein into an abnormal oligomer constitutes a general mechanism by which missense mutations rewire cellular signalling pathways in cancer.

Funded by ERC (StG-242958; MMM), Utrecht University (High Potential; MMM & SGDR), MRC (U117584268; JPV), EMBO (983-2009; HN), Uehara and Kanae Foundations (HN).

S7-02 Wnt signaling in the annelid *Platynereis dumerilii*

Demilly, Adrien (Institut Jacques Monod CNRS, Paris, FRA); Gazave, Eve (Institut Jacques Monod CNRS, Paris, FRA); Steinmetz, Patrick (University of Vienna, AUT); Marchand, Lauriane (Institut Jacques Monod CNRS, Paris, FRA); Kerner, Pierre (Institut Jacques Monod CNRS, Paris, FRA); **Vervoort, Michel** (Institut Jacques Monod CNRS, Paris, FRA)

Wnt genes encode signaling molecules that regulate a wide array of developmental processes including the formation of the nervous system. Wnt molecules signal through several different intracellular pathways including the beta-catenin and Planar Cell Polarity (PCP) pathways. In vertebrates for example, a gradient of Wnt ligands produced by the roof plate stimulates both proliferation and differentiation of neural progenitors in a context-dependent manner, through the activation of the Wnt/beta-catenin pathway. Other Wnt molecules, mainly acting through the PCP pathway, also participate in key events of vertebrate CNS development such as neural tube closure, floor plate formation and axon guidance. We studied Wnt/beta-catenin and Wnt/PCP signaling during the development of the polychaete annelid *Platynereis dumerilii*. Over the past decade, *Platynereis* has become a valuable model for evolutionary developmental biology studies, especially regarding the evolution and development of the nervous system and segmentation. We have cloned and analyzed the expression of several genes that encode members of the Wnt/beta-catenin and Wnt/PCP pathways in *Platynereis*. We also analyzed the functions of these pathways using pharmacological inhibitors. We will in particular show that Wnt/beta-catenin is required for the transition between proliferating neural progenitors and differentiating neurons during the development of the central nervous system. We will also report expression and functional data suggesting an involvement of the Wnt/ PCP pathway in nervous system morphogenesis. Finally, we will discuss how these data may help to better understand the evolution of Wnt gene functions in bilaterians.

S7-03 Wnt signaling shapes planarians

Adell, Teresa (University of Barcelona, ESP); Sureda, Miquel (University of Barcelona, ESP); Almuedo-Castillo, Maria (University of Barcelona, ESP); Martín-Durán, José María (University of Barcelona, ESP); Rojo-Laguna, Jose Ignacio (University of Barcelona, ESP); Saló, Emili (University of Barcelona, ESP)

Planarians are a classical model for regeneration studies due to their unique plasticity. They can regenerate a whole animal from any piece of their body and show a continuous remodeling. This capacity relies on the pluripotency of their adult stem cells, which must be accompanied by the continuous activation of major developmental signaling pathways to control their proliferation and fate. Despite its broad range of functions, the ß-catenin-dependent Wnt signaling is a conserved mechanism to pattern the antero-posterior axis during embryogenesis across metazoans. The ß-catenin-independent Wnt signaling includes unrelated branches, involved in the regulation of different processes such as planar cell polarization (PCP), cell division orientation, ciliogenesis or neural circuitry assembly. In adult planarians, both during regeneration and homeostasis, silencing of ßcatenin1 produces a gradual anteriorization that ends with a striking fully anteriorized radial-like planarian. Out of the nine Wnts that integrate the Schmidthea Wnt family, four of them are expressed in the posterior part of the animal, being a Wnt1 homolog the main ß-catenin1 activator. Moreover, secreted Wnt inhibitors are found in the most anterior planarian tip. These data suggest that a graded activation of ß-catenin1 underlies planarian antero-posterior identity specification. However, using a specific anti-ß-catenin1 antibody, we found that ß-catenin1 does not show a graded activation but it is broadly nuclearized all around planarians. Moreover, during regeneration it is activated in both anterior and posterior regenerating blastemas as well as in any regenerating organ. These results prompt us to propose a model in which a local activity rather than a general axial gradient of ß-catenin1 would pattern planarian antero-posterior axis. Finally, two ß-catenin-independent Wnt branches have been functionally analyzed in planarians: a Wnt-independent one, integrated by Dishevelled, Van-Gogh and Diversin, which is required for apical positioning of the cilia basal bodies of planarian epithelial cells; and a Wnt5-dependent one, which controls the proper medio-lateral positioning of the central nervous system. Here we show that a Ror-family receptor tyrosine kinase is the responsible for receiving the Wnt5 signal in planarians. Our current working model proposes that Wnt5 and Slit, a conserved repulsive axonal cue, integrate a self-regulated system to define the path of the nervous projections through restricting their expression boundaries.

S7-04 Dissecting Wnt-signaling dependent transcription in the mouse

Cantù, Claudio (University of Zurich, CHE)

The Wnt-signaling pathway plays a major role in virtually all developmental processes, as well as in the homeostasis of adult tissues. The aberrant activation of this pathway is associated with cancer formation and progression. The Wnt/ß-catenin dependent transcription has been proposed to be promoted, at least in Drosophila, via a "chain of adaptors". In this model, the four proteins — Pan/TCF > Arm/ß-catenin > Lgs/Bcl9>Pygo — serially recruit each other to the DNA in order to efficiently activate Wnt-target gene expression (reviewed in Mosimann et al. 2009). According to this model Lgs simply serves as an adaptor protein to recruit Pygo to Arm transcriptional complex. However, the relevance of the "chain of adaptors" during mouse development remains largely unexplored. In mice there are two lgs, Bcl9 and Bcl9l (Bcl9/9l), that bind Pygo and ß-catenin via two evolutionarily conserved domains, HD1 and HD2, respectively. In order to dissect the relevance of these two interactions, we generated knock-in mouse strains in which these domains are deleted. We find that the individual abrogation of both interactions causes embryonic lethality, underscoring the physiologica lrelevance of their integrity. Importantly, we also findthat the Bcl9/9l-Pygo axis is required for lens development: while lens formation is critically dependent on the presence of the HD1 domains, it is not affected by the lack of the HD2 domains, indicating that Bcl9/9l act in this context in a Pygo-dependent, but ß-catenin-independent manner. Furthermore, we uncover the existence of a new regulatory circuit: the Bcl9/9l-Pygo complex is required for the early expression of Pax6, the master regulator of eye development; in turn, Pax6 directly activates Bcl9/9l transcription, possibly providing a positive feedback loop that sustains its expression. We envision that the genetic models we generated might provide insights to settle the debate on the Wnt-signaling dependent and independent roles of Bcl9 and Pygo, and shed light on their mechanism of action in both scenarios.

14.00 – 15.40 Symposium S8: Quo vadis EvoDevo?

ROOM C2

Organizers: Manfred Laubichler and Cassandra Extavour
Chair: Manfred Laubichler
Discussion leader: Ronald Jenner

S8-01 EvoDevo in the Americas: A report on a community workshop to consolidate and advance the field of Evolutionary Developmental Biology

Extavour, Cassandra (Harvard University, Cambridge, MA, USA)

[No abstract available]

S8-02 The future of evo-devo

Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

[No abstract available]

S8-03 Quo vadis EvoDevo?

Laubichler, Manfred (Arizona State University, Tempe, USA)

The field of Evo Devo is currently at a crossroads. We have many well-established approaches that continue to generate data but are no longer contributing much to the conceptual and theoretical excitement that has characterized earlier periods in the history of EvoDevo. At the same time the field continues to struggle with a number of conceptual issues that have not been resolved, such as the integration of stander ovulation genetic approaches into an EvoDevo framework (or vice versa), the evo devo or devo evo split, the inclusion of ecological perspectives or the emergence of synthetic and computational approaches. All this conceptual uncertainty has also contributed to an institutional and funding "crisis" (at least in the US). In response to this a working group at NESCent has been bringing together a broad array of EvoDevo researchers (from all career stages with a bias towards the younger generations and largely, but not exclusively North America based) to discuss future directions of the field. In this session we would like to introduce a number of the issues raised by the NESCent group and start a dialogue with our European colleagues. We envision a set of paired talks, with one person presenting a review/report of a NESCent discussion and a European colleague presenting a response. This way we hope to foster a dialogue between the two communities and lay the groundwork for a more intensive collaboration in the future (one of the goals of the NESCent group).

16.10 – 17.10 **Contributed Session C6:**
Plant EvoDevo: Linking cross-species genetic and morphological variation

ROOM A

Chair: Caspar Chater

C6-01 Deep homology in the land plant stomatal development programmed

Chater, Caspar (University of Sheffield, GBR); Caine, Robert (University of Sheffield, GBR); Kamisugi, Yasuko (University of Leeds, GBR); Cuming, Andrew (University of Leeds, GBR); Fleming, Andrew (University of Sheffield, GBR); Beerling, David (University of Sheffield, GBR); Gray, Julie (University of Sheffield, GBR)

Stomatal development in angiosperms is controlled by a suite of positive and negative regulators of cell development. Stoma-like complexes are found on ancient plant cuticle fossils and on the sporophytes of extant mosses and hornworts. It is not known, however, whether these are orthologous structures to tracheophyte stomata. Recent evidence suggests angiosperm-like molecular signalling and physiological responses in the stomata of the model moss *Physcomitrella*. Here we present evidence that PpSMF1, a moss gene homologous to Arabidopsis bHLH transcription factors SPEECHLESS, MUTE, and FAMA, that regulate stomatal formation in the moss. Strikingly, moss sporophytes lacking PpSMF1 expression produce no stomata. This strongly suggests an ancient origin of stomata and deep homology in the stomatal development tool kit across all extant land plants. As well as conservation of these positive stomatal regulators, we also show that negative regulator PpEPF1, homologous to EPIDERMAL PATTERNING FACTORS 1 AND 2, and the stomatal modulator TOO MANY MOUTHS (TMM) act to control stomatal patterning in the moss, as they do in Arabidopsis. Our data therefore strongly suggest that the core machinery for stomatal development is conserved across land plant evolution and that all extant stomatous lineages share a common ancestor that lived over 410 mya.

C6-02 Through the periscope: Understanding early grass leaf development

Richardson, Annis (John Innes Centre, Norwich, GBR); Rebocho, Xana (John Innes Centre, Norwich, GBR); O'Connor, Devin (Cambridge University, GBR); Hake, Sarah (University of California Berkeley, CA, USA); Coen, Enrico (John Innes Centre, Norwich, GBR)

The plant kingdom exhibits huge diversity in leaf morphologies and shapes, often influencing an individual's fitness. An excellent example of shape adaptation in leaves is found in the grasses. The grass leaf is periscope-like which enables the meristem to remain at the base of the plant protected from herbivore grazing until flowering, whilst still able to gain height in the leaves to compete with neighbours for light. But how does this wrapped, periscope-like leaf structure develop? Initial morphological studies using published data and optical projection tomography (OPT)(1) identified several key morphological transitions from a ring, to a hood, to a more cone-like primordium. In addition these studies highlighted two phases of growth; the first arching of the ring to form a hood and the second more vertical growth of the hood to form a cone-like shape. To explore hypotheses relating to these early stages in grass leaf morphogenesis we used Growing Polarised Tissue framework (GPT-framework (2)) modelling. Through modelling we have identified a double switch in tissue cell polarity and growth rate pattern that can account for these morphological transitions during early grass leaf development. I will present the current model along with in planta data that support this double-switch hypothesis. The double switch in tissue cell polarity and growth rate pattern could be key to the evolution of the periscope-like grass leaf.

C6-03 Three ancient hormonal cues co-ordinate shoot branching in a moss

Coudert, Yoan (University of Cambridge, GBR); Palubicki, Wojtek (Cambridge University, GBR); Leyser, Ottoline (Cambridge University, GBR); Harrison, Jill (University of Cambridge, GBR)

Branching patterns are a primary determinant of plant architecture and strongly impact on productivity by regulating light harvesting potential and resource allocation. Plants colonized land over 450 million years ago and underwent architectural diversification in the haploid and diploid stages of the life cycle independently. Although similar branching mechanisms evolved in both life cycle stages, our functional understanding of branching is limited to diploid flowering plant models such as *Arabidopsis*. To test whether the same molecular cues regulate branching mechanisms that have evolved convergently, we undertook a computational and genetic analysis of branching patterns in the haploid leafy shoot of a moss. We show that a simple model co-ordinating the activity of shoot tips across the plant can account for the branch distribution, and that three known hormonal regulators of branching in flowering plants generate the pattern. Importantly, these cues have been independently recruited during evolution to regulate branching patterns in both haploid and diploid life cycle stages, and may be integrated via a novel mechanism in moss.

C6-04 Stay high or get low: Can epigenetic variation lead to recurrent speciation?

Paun, Ovidiu (University of Vienna, AUT); Flatscher, Ruth (University of Vienna, AUT); Frajman, Bozo (University of Innsbruck, AUT); Trucchi, Emiliano (University of Vienna, AUT); Schönswetter, Peter (University of Innsbruck, AUT)

Across heterogeneous environments, variation in biotic and abiotic conditions and in the resulting selective pressures often leads to the formation of ecotypes, i.e., distinct populations adapted to their specific habitat. Whereas they are usually still inter-fertile with other conspecific ecotypes, limited gene flow may over time lead to speciation. An interesting example for recurrent independent formation of ecotypes is found in the species complex of the Southeastern European mountain plant *Heliosperma pusillum* (Caryophyllaceae). Within this group, the lower elevation *H. veselskyi* is a perennial species with a dense sticky indumentum, which inhabits rock overhangs and shallow caves with poor light conditions below the timberline, whereas its closest relative, the alpine *H. pusillum*, is glabrous and occurs on creeks and moist calcareous screes. This morphological differentiation remains stable in offspring grown from seeds in a common garden at least across two generations. Reciprocal transplantations show that *H. pusillum* has higher germination rates in both habitats, but only in the alpine one its growth parameters are higher than for *H. veselsky*. Although they have been both described at species rank because of their distinct morphology, molecular data (for example over 2,200 informative SNPs filtered from NGS-based restriction site associated DNA sequencing — RADseq) surprisingly show that the two species are intermixed, suggesting a recent and recurrent origin of *H. veselskyi* from *H. pusillum*. Moreover, individuals of both ecotypes stemming from one mountain range are genetically more similar than individuals of the same ecotype from different mountain ranges. This suggests that they are ecotypes, resulting from middle- to short-term adaptive processes, perhaps under the influence of the environment and independent (yet!) of actual changes in DNA sequence. Genomic scans of genetic data performed with BayeScan have confirmed the complete absence of any genetic outlier loci that could have been involved in the phenotypic differentiation between the two taxa. We present preliminary results based on bisulfite RADseq to test for genome-wide differences in DNA methylation correlated with the striking phenotypic differentiation between high- and low altitude populations and discuss here the possible role of epigenetics in the initial phase of divergent evolution.In the light of our results a likely evolutionary scenario for this group appears to involve environmental disruption of gene expression levels, which have independently converged to similar, stable phenotypes through repeatable effects of natural selection in similar environments.

16.10 – 17.10 Contributed Session C7: EvoDevo of symmetry in animals and plants

ROOM B

Chair: Sophie Nadot

C7-01 A computational model of cleavage patterns in metazoa and its evolution

Brun-Usan, Miguel (Universitat Autònoma Barcelona, Bellaterra, ESP); Salazar-Ciudad, Isaac (EvoDevo Helsinki Community, FIN)

Cleavage is the developmental stage whereby the fecunded oocyte gives rise to a cluster of cells (blastomeres) with a particular spatial pattern and with few or no cell growth. In general, cleavage takes place inside a spheroidal hyaline layer with constant volume and it involves rapid cell divisions. Different cleavage patterns are found among metazoans, but all of them fit into a small set of basic patterns. However these patterns do not seem to follow an evident phylogenetic relationship. During cleavage, dividing cells are oriented either by the longest axis of the cell (Hertwig's rule), or by external cues (a morphogen gradient or the contact area with neighbouring blastomeres). Cell shape is, in turn, affected by adhesion strenghts, surface tension and packing constraints. Moreover, some cell internal assymetries result in unequal cell divisions and the amount of yolk diminishes cell division rate. How these processes are spatio-temporally coordinated to generate the observed diversity in cleavage patterns remains unclear. In order to assess this question, we have developed a computational model capable reproducing all the cell behaviors (division, adhesion, contraction, signaling, matrix secretion, etc.) that blastomeres use. By using this model, our aim is to build a theoretical generative morphospace of all possible cleavage patterns. By comparing it with biological data, we measure which proportion of the morphospace has been explored by metazoan and which has not. Moreover, our model can shed light on which are the more likely patterns, and which evolutionary transitions between cleavage patterns are easier to occur. In the case of the spiralian pattern, we have found that some physical (non-genetic) interactions such as cell-surface tension and packing constraints suffice to explain a large part of the symmetry breaks of this cleavage pattern.

C7-02 The establishment of left-right asymmetry during spiralian development in the serpulid annelid *Pomatoceros lamarcki*

Namigai, Erica (University of Oxford, GBR); Shimeld, Sebastian (University of Oxford, GBR)

The establishment of body axes is a fundamental process of metazoan development. However, how the left-right (LR) axis is established remains poorly understood, especially in organisms outside of the Deuterostomia and Ecdysozoa. I am studying the mechanisms underlying LR asymmetry establishment in the Lophotrochozoa using *Pomatoceros*

lamarcki, a serpulid annelid, as a model. To observe the first instance of symmetry breaking, we are using live imaging to understand biases during axis establishment at early cleavage stages, including the cellular architecture and dynamics of early spiral cleavage. We have also sequenced the *P. lamarcki* genome, and key genes with a conserved function in LR asymmetry (i.e., Nodal) are being analyzed. This research addresses key questions concerning body axis establishment during spiralian development, and is valuable for exploring the dynamic aspects of early development and to understand the evolution of LR asymmetry within the Lophotrochozoa.

C7-03 Colony symmetry in thecate hydroids (Cnidaria, Hydroidomedusa, Leptomedusae): Transition from radial to bilateral symmetry

Kosevich, Igor (Lomonosov Moscow State University, RUS)

Soft tissue morphology and organisation of certain elements (modules) in colonies of thecate hydroids evidently display the predominance of radial symmetry. At the same time, the skeleton structures of shoot modules and entire shoots of the colonies demonstrate either absence of predominant type of symmetry or availability of different combinations of bilateral and translational symmetry (glide reflection symmetry, helical symmetry, etc.). The only colony module that is evidently bilateral is the stolon tip growing over the substrate. In most cases, the outer organisation of the morphogenetic shoot modules in thecate hydroids also displays bilateral symmetry. There is one plane of symmetry, so it is possible to mark out left and right, adjacent and opposing ("dorsal" and "ventral") sides of the module. The only module that possesses radial symmetry is the primary module developed from the settled larva. However, emerging secondary modules switches to bilateral symmetry that determines the shoot organisation. Transition to the bilateral symmetry of the morphogenetic modules in thecate colonial hydroids can be explained by the model of growth and development regulation in unitary and modular hydroids proposed by Berking (2003, 2006; Berking & Herrmann 2010) and based on the idea of positional information (Wolpert 1971, 2011). The primary module developed from settled larva possesses radial symmetry. Development of its parts proceeds according to the positional information increasing during growing tip activity. The secondary growing tip emerging on the primary module is functionally bilateral starting from the origin: the adjacent to the parental hydranth side has higher value of positional information compared to opposing one. Henceforward all the secondary shoot and stolon modules are bilateral. Functional bilateralism stabilises morphogenesis in colony shoots providing regular spatial organisation without any need in new mechanism of morphogenesis regulation.

C7-04 Evolution of a novel flower trait in the Brassicaceae

Busch, Andrea (University of Osnabrück, GER); Horn, Stefanie (University of Osnabrück, GER); Zachgo, Sabine (University of Osnabrück, GER)

A relevant step in angiosperm radiation was the adaptation to pollinators via the establishment of flower monosymmetry, which evolved repeatedly in different angiosperm lineages. In all taxa analysed so far, monosymmetry development is controlled by TCP transcription factors. The Brassicaceae are dominated by polysymmetric species and only six genera form flowers with two petal pairs of different sizes, making it an ideal system to study monosymmetry evolution. In *Iberis amara*, the first monosymmetric crucifer analysed, unequal petal pair formation is due to a stronger expression of *IaTCP1* in the smaller, adaxial petals. We show that this is also the case for additional *Iberis* species and two other monosymmetric crucifer genera. A phylogenetic reconstruction of the crucifers places all monosymmetric species in one clade. Analyses of the early and late expression of *TCP1* orthologs in mono- and polysymmetric members, which are representative of the four main crucifer lineages demonstrate that crucifer monosymmetry evolved via a heterochronic expression shift from an early adaxial expression in floral meristems in polysymmetric species to a late expression in adaxial petals in monosymmetric members. Using RNAseq and Arabidopsis microarray technology we obtained an overview of the *I. amara* petal transcriptome to allow a closer insight into the molecular network controlled by *IaTCP1*.

16.10 – 17.10 Contributed Session C8: Perspectives on Wnt signaling I

ROOM C1

Chair: Reinhard Schröder

C8-01 A Wnt landscape regulates segment polarity in the annelid *Platynereis*

Balavoine, Guillaume (Institut Jacques Monod, Paris, FRA); Gazave, Eve (Institut Jacques Monod / CNRS, Paris, FRA)

The debate on the origin of segmentation is a central question in the study of body plan evolution in metazoans. Annelids are the most conspicuously metameric animals as most of the trunk is formed of identical anatomical units. We are studying segmentation processes in the marine annelid, *Platynereis dumerilii*. Segments are formed in two consecutive waves: three anterior-most larval segments are formed simultaneously when the unsegmented trochophore larva metamorphoses into a small worm and later, a sub-terminal posterior segment addition zone produces all the other segments sequentially throughout the worm's life. We studied the larval and juvenile expression patterns of the twelve Wnt ligands existing in *Platynereis*. Half of these ligands

are expressed in banded patterns that suggest involvement in segment polarity. The patterns are complementary along the segment axis, suggesting that a form of combinatorial signalling or "Wnt landscape" is at play to shape the segment. We test the role of the main pathway regulated by Wnts, beta-catenin, through the use of small molecule agonists and antagonists. Both sets of treatments give small worms with highly abnormal undifferentiated segments. Remarkably, Arthropods show similar expressions of Wnt genes in segment primordia suggesting that segment polarity is an ancient role of Wnt signalling inherited from a protostome annelid-like ancestor.

C8-02 Functional analysis of Wnt signalling in early spider development shows diverse roles in embryonic patterning

Eibner, Cornelius (Friedrich Schiller University Jena, GER); Pohl, Kerstin (Friedrich Schiller University Jena, GER); Beyerlein, Anna (Friedrich Schiller University Jena, GER); Holzem, Michaela (Friedrich Schiller University Jena, GER); Damen, Wim GM (Friedrich Schiller University Jena, GER)

We study the role of canonical Wnt signalling in the development of the spider Parasteatoda tepidariorum by the knockdown of Wnt pathway components via RNAi. Using Pt-axin RNAi or Pt-armadillo/beta-catenin RNAi we are able to up-regulate or inhibit Wnt signalling, leading to diverse phenotypes. These phenotypes, including segmentation and axial phenotypes, partly show resemblance of earlier reported Wnt8-RNAi phenotypes in Parasteatoda. Phenotypes that are different from the Wnt8-RNAi phenotype indicate that other Wnt genes also play important roles during early embryonic development, especially in axis formation and segmentation. This is further corroborated by expression patterns of other Wnt genes. Wnt signalling plays a key role in two very important processes: antero-posterior axis formation and segmentation. It can be assumed that this is not entirely directed by one Wnt gene but that there is everything from a strict division of tasks to a combinatorial way of action. We investigate the interactions of Wnt signalling with other posteriorly expressed genes like caudal, which is expressed dynamically in Parasteatoda, and we look at the expression of Wnt antagonists like FGF and axin and their role in shaping an antero-posterior Wnt gradient. We further present first data from a Parasteatoda transcriptome and the genome and discuss the complexity of Wnt signalling in spiders. The spider Parasteatoda possesses homologues of 12 Wnt genes from 10 families, five of which are shared with Drosophila and nine with human. Members of the Wnt3, Wnt9 and Wnt10 families have been lost in Parasteatoda. We also analyse the complexity of other Wnt associated pathways like the hippo pathway. We think that the accessibility to functional tools, genomic resources, its phylogenetic position and the large complement of Wnt signalling components suggest that Parasteatoda has the potential to advance to become one of the main non-vertebrate model system, besides Drosophila.

C8-03 Characterization of the gene regulatory network for posterior development in the spider *Parasteatoda tepidariorum*

Schoenauer, Anna (Oxford Brookes University, GBR); Schwager, Evelyn (Oxford Brookes University, GBR); Hilbrant, Maarten (Oxford Brookes University, GBR); Damen, Wim G. M. (Friedrich Schiller Universität Jena, GER); McGregor, Alistair P. (Oxford Brookes University, GBR)

Short germ arthropods form most of their body segments from a posterior segment addition zone (SAZ) in a sequential fashion. There is increasing evidence for spiders and other arthropods that Wnt and Delta-Notch signaling regulated the formation and function of the SAZ ancestrally in arthropods, analogously to the regulation of somitogenesis in vertebrates. However, it remains unknown how these signaling pathways interact with other putatively downstream genes, such as *even-skipped*, in arthropods that employ Wnt and Delta-Notch signaling for segmentation. To answer this question, we characterized the expression of the pair rule gene orthologue *even-skipped (Pt-eve)* during posterior development in *Parasteatoda tepidariorium*. In this spider, *Pt-eve* exhibits dynamic expression in the SAZ that resolves into stripes of expression in nascent segments. To then study the role and regulation of *Pt-eve* during embryogenesis, we investigated the effect of *Pt-Wnt8* and *Pt-Delta* parental RNAi knockdown on *Pt-eve* expression. In situ expression analysis showed that *Pt-eve* is surprisingly still expressed in a dynamic pattern in *Pt-Wnt8* RNAi embryos, suggesting that *Pt-Wnt8* does not directly regulate *Pt-eve*. This *Pt-eve* expression putatively overlaps with *caudal (Pt-cad)* expression domains in *Pt-Wnt8* RNAi embryos. In contrast, in *Pt-Delta* RNAi embryos, we observed that *Pt-eve* is no longer expressed in the SAZ, but is still observed in stripes in one or two nascent segments. Assuming that the establishment of the SAZ and the formation of segments from this tissue are independently regulated, we are currently examining the regulatory interactions between *Pt-cad* and *Pt-eve* in the SAZ and the nascent segments. By injecting *Pt-cad* dsRNA into single cells, leading to an RNAi effect only in a small clone area we found that Pt-eve is regulated by *Pt-cad* in the SAZ and we are presently investigating the regulation of *Pt-eve* in the forming segments.

C8-04 Anterior/posterior patterning of the dorsal mesoderm in vertebrates evolved as a novelty from the ancestral chordate mesoderm by a heterotopic shift

Onai, Takayuki (RIKEN Center for Developmental Biology, Kobe, JPN); Aramaki, Toshihiro (Osaka University, Osaka, JPN); Inomata, Hidehiko (RIKEN Center for Developmental Biology, Kobe, JPN); Hirai, Tamami (RIKEN Center for Developmental Biology, Kobe, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

To elucidate the evolutionary origin of anterior/posterior (A/P) patterning system of the vertebrate dorsal mesoderm, we investigated the molecular architecture of A/P patterning of the dorsal mesoderm in amphioxus and vertebrates. Here, we report that A/P patterning mechanism of the vertebrate dorsal mesoderm is fundamentally different from amphioxus. A/P progenitor domains Gsc, Delta and Brachury are initially overlapped in amphioxus and vertebrates. These domains heterotopically shift antero-posteriorly in vertebrates but remain overlapped in amphioxus. Suppression of the heterotopic shift by arrest of cell migration in vertebrates recapitulates the amphioxus patterning. The Wnt/β-catenin signal gradient and cadherin degradation system that regulate A/P patterning in vertebrate are missing in amphioxus. Our data indicate that A/P patterning of the dorsal mesoderm in vertebrates evolved by a heterotopic shift of domain genes from ancestral chordates. We propose the genetic mechanism of heterotopic shift as a common constraint to increase the morphological complexity in evolution.

16.10 – 17.10 Contributed Session C9: Mechanical mechanisms of development

ROOM C2

Chair: Derek Moulton

C9-01 The 3D Crocs project: Physical mechanisms generate a diversity of cranial scale patterns among Crocodylia

May, Catherine (University of Geneva, CHE); Milinkovitch, Michel (University of Geneva, CHE)

Species of Crocodylia have thick, armored skin that is paradoxically also very sensitive. For example, the face and jaw scales of Crocodilians might have higher mechanical sensitivity than human fingertips due to highly branched nerve networks associated with many micro-organs, referred to as "integumentary sensory organs" (ISOs). Recently, we demonstrated that ISOs are multi-sensory as they exhibit a combined sensitivity to mechanical, thermal and chemical stimuli (Di-Poi and Milinkovitch 2013), with no equivalent in the sensory systems of other vertebrate lineages. It has long been known that skin appendages such as hair, feathers, and scales form from developmental units (primordia) during embryogenesis. However, we recently discovered that the polygonal head scales of crocodiles do not form from such genetically determined units, but are generated through a physical mechanism: the cracking of living tissue in a stress field (Milinkovitch et al. 2013). Cracks in the developing skin propagate to form a network, leaving polygonal domains of skin corresponding to head scales. Here, we further investigate the developmental-physical phenomenon that drives scale-cracking patterns among different species of Crocodylia. Using high-resolution photographs and multi-view stereo approaches, we generated 3D surface reconstructions of the head of various alligator, caiman, gavial and crocodile species. Next, scale features such as edges, nodes and ISOs were marked on virtual 3D mesh models. We analyze the distribution of ISOs to identify how they may contribute to surface topology, such as constraining the spatial propagation of cracks. Lastly, the topology and geometry of head scale patterns were quantitatively analyzed to investigate the parameters that drive natural variations in skin surface cracking patterns among different Crocodilian species.

C9-02 Epithelial cell shaping in response to mechanical cues is an evolutionary conserved way for sculpting an embryo

Kraus, Yulia (Lomonosov Moscow State University, RUS); Kremnyov, Stanislav (Lomonosov Moscow State University, RUS); Frank, Uri (National University of Ireland, Galway, IRL)

To a great extent, embryonic shape is a product of epithelial sheets morphogenesis based on coordinated cell shape changes and cell movements (Davidson 2012). The role of mechanical cues modulating the morphogenesis deserves increasing interest. However, we know very little about the evolutionary basal reactions of epithelial cells to mechanical stimuli. Comparative analysis of cell behavior in basal metazoans and in bilaterians may be instructive in understanding how these reactions evolved. The cnidarian representative, *Hydractinia echinata*, is a model organism for developmental biology. Gastrulation in *Hydractinia* proceeds by mixed delamination and gastrulating embryo lacks any morphological sign of the primary body axis. However, there is an absolutely unique landmark of embryonic polarity that we described as a gradient of order. The maximum of order coincides with the future anterior pole of the larva, and the most morphologically disordered region of an embryo is the future posterior pole. The posterior half of an embryo is covered by multiple ectodermal folds, with bottle cells located on the concave side of the fold. Formation of bottle cells and ectodermal folds has no known function in *Hydractinia* development, and we propose that these are the side effects of rapid cell proliferation in the ectoderm leading to an increase of planar compression in epithelial sheet. We assume that compressed ectodermal cells start

to reduce the apex area in order to reduce compression and restore normal tension. Gradual constriction of apices of neighboring cells automatically leads to macromorphological consequence — bending of the epithelium and formation of folds. Notice that *Wnt3* transcripts form a countergradient to the gradient of order and can facilitate the morphogenesis in the posterior region. Experimental support for our assumptions can be derived from data obtained on the explants of the amphibian embryo. We have shown that the explant of the blastocoel roof from early gastrula actively reacts to mechanical compression produced by an ectopically applied force by apical constriction of cells acquiring the bottle shape (Kremnyov et al. 2012). Interestingly, the reaction of cells depended on the explant origin. This indicates that cells that are strictly programmed to perform important morphogenesis (such as formation of neural folds) are able to resist accidental mechanical stimuli. In conclusion, epithelial cells respond to mechanical cue in a manner of self-organization, as in both model systems this response had no developmental function. The capability of regulation of unbalanced mechanical stresses by active cell shaping is an intrinsic characteristic of metazoans' epithelial cell. This ability can be tuned in evolution under imposed developmental constraints.

C9-03 Comparative tissue dynamics of late embryogenesis in flies and midges

Fraire-Zamora, Juan J. (Centre de Regulacio Genomica, Barcelona, ESP); Solon, Jerome (Centre de Regulacio Genomica, Barcelona, ESP); Jaeger, Johannes (Centre de Regulacio Genomica, Barcelona, ESP)

The development of a multicellular organism can be viewed as a series of spatial reorganization events that follow established tissular genetic patterns. A recent renaissance of the mechanical view of development has pointed out the action of mechanical forces (such as pulling, twisting, buckling and/or bending) on tissue remodelling and their tight interplay with the genetic processes that orchestrate morphogenesis. This view motivates exciting questions on whether physical constraints play a role in the evolution of developmental processes or if the evolution of gene network results in specific physical properties of tissues. Our research project focuses on the comparative study of tissue dynamics and cellular forces that shape dipteran embryos of *Clogmia albipunctata*, *Megaselia abdita* and *Drosophila melanogaster*. We aim to establish an interdisciplinary experimental approach to obtain comparative maps of embryonic mechanical forces that can be linked to gene expression. We are currently exploring methodologies that allow us to obtain time-lapse sequences of dipteran embryo development and combining them to traditional immunostaining and in situ hybridization techniques to understand the genetic and phenotypic differences that lead to completion of development. For this meeting, we will present fluorescence time-lapse sequences of *M. abdita* embryos undergoing dorsal closure and compare them to *D. melanogaster*. We will also present our results on the comparison of contractile structures (i.e., actin cables and extraembryonic tissues) that lead to dorsal closure in both dipteran species. We envision that our experimental approach will reveal new insights in the interplay of mechanics and genetics during the evolution of development.

C9-04 Shaping the snapdragon's mouth using a CUP

Rebocho, Xana (John Innes Centre, Norwich, GBR); Ableу, Katie (John Innes Centre, Norwich, GBR); Bradley, Desmond (John Innes Centre, Norwich, GBR); Copsey, Lucy (John Innes Centre, Norwich, GBR); Bagham, Andrew (University of East Anglia, Norwich, GBR); Coen, Enrico (John Innes Centre, Norwich, GBR)

The evolution of organ shape is deeply connected with the function of the organ. Flowers evolved their shapes, colors and smells to attract their pollinators and the Antirrhinum flower is a beautiful example of co-evolution with the bumblebee. The zygormorphic Antirrhinum flower has a complex 3D shape that arises through the deformation of the petals to form a close mouth flower, a morphological novelty that may have led to a pollinator shift and reproductive isolation. Recent studies in our laboratory, using a combination of molecular genetics, clonal analysis and computational modelling, have provided a working hypothesis of how seemingly complex shapes, such as that of the Antirrhinum flower, can be generated through an interplay between polarities and identities. In particular, the model predicts that the lower petal deformation arises within the palate and lip regions by a change in the pattern of anisotropic growth oriented by a tissue cell polarity reorientation. We found that CUPULIFORMIS (CUP), the homologue of Arabidopsis CUC1 and CUC2, is expressed in the palate and lip regions and initiates as a narrow band in between the tube and lobe enlarging during petal development. CUP is necessary for the growth of these regions as in cup mutants these regions fail to develop resulting in a simpler flower shape (with an open mouth). To test if there is a reorientation of polarity within the palate and lip as postulated in the model, we visualised the PIN1 polarity field (a plant cell polarity marker) in developing petals using whole mount immune localization. We observed a local reorientation of the PIN1 cell polarity particular to the ventral petal junctions that are under the control of CUP. The detailed analysis of the expression pattern of CUP and the cellular behaviour of PIN1 suggest a genetic and cellular mechanism for the formation of a 3D tissue deformation based on a feedback between gene identity and tissue cell polarity. Our results suggest that the re-deployment of a boundary gene during petal development may have played a role in the evolution of the 3D shape of the Antirrhinum flower.

16.10 – 17.10 **Contributed Session C10:**
Complexity in gene networks and structures

ROOM D

Chair: Andrew Cridge

C10-01 Constraining and buffering: Regulation and evolution of complex gene networks

Cridge, Andrew (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL)

Our aim is to understand how conserved genes change their role in the evolution of the insect segmentation network, how the network buffers change, and how that might constrain, or confer diversity of body plan. Antibodies to the early sementation transcription factors *caudal (cad), hunchback (hb)* and *orthodenticle (otd)* were developed for *Drosophila melanogaster*, *Apis mellifera* and *Acyrthosiphon pisum*. These antibodies were tested via immunohistochemistry and immunoblotting to find the most specific for Chromatin Immunoprecipitation (ChIP). ChIP was performed on embryonic tissue and the isolated genomic DNA was sequenced using next-generation sequencing technology. The sequence reads were mapped back to their respective genomes to identify regions enriched for transcription factor binding. These regions were analyzed to identify unique *cis*-regulatory motifs (CRMs) and regulated genes. The ChIP-seq study identified multiple *cis*-regulatory motifs (CRMs) and gene targets for the three transcription factors in each of the three insect species studied. Bioinformatic analysis allowed confident prediction of biologically important CRMs, as well as providing information on the binding specificity of the transcription factors. This highlighted how the genes regulated by these transcription factors have changed over evolutionary time. This enabled us to identifying the core evolutionary functions that these genes carry out in all three insects, and their ancestral roles in insect segmentation. We also identified genes that are regulated in only one species. These genes represent mechanisms that buffer changes in the regulation of our key transcription factors thus allowing the conserved segmentation output. By continuing to study these genes we will learn how genes become co-opted into developmental networks, how such co-opted genes integrate with the rest of the network, and if these genes act to buffer regulatory changes in the transcription factors themselves. This data has provided us, for the first time, with an understanding of how the targets of key transcription factors change over evolutionary time, effectively a measure of evolutionary change in a complex transcription factor network.

C10-02 Structure, function and development of an extraordinary insect eye

Buschbeck, Elke (University of Cincinnati, OH, USA); Stahl, Aaron (University of Cincinnati, OH, USA); Cook, Tiffany (Cincinnati Children's Hospital Medical Center, OH, USA)

Sunburst Diving Beetle (*Thermonectus marmoratus)* larvae are highly specialized, visually guided predators. Their primary eyes are uniquely organized single-chamber eyes, with bifocal lenses, multiple retinas, polarization sensitivity and sophisticated distance vision, raising the question of how this bizarre eye organization may have arisen developmentally. Data on eye development is nearly exclusively available for the image-forming camera eyes of vertebrates (primarily mice) and the compound eye of insects (primarily *Drosophila*). Despite these phylogenetically and functionally diverse systems, common gene networks have started to be recognized between both eye types, leading to the hypothesis that key components of these networks also give rise to other strongly divergent eye organizations such as those of *Thermonectus* larval eyes. To test this hypothesis we obtained transcriptomes of *Thermonectus* head tissue, and identified many key eye-development genes. These include genes associated with eye specification (e.g., *twin of eyeless (toy)*, *eyes absent (eya)*, and *sine oculis (so)*); photoreceptor specification (e.g., *orthodenticle, BarHI, homothorax, spalt* and *sevenless*); photoreceptor function (e.g., *rhodopsins, arrestin, crumbs* and *prominin*); and lens development (e.g., *prospero, D-pax2* and *cut*). A detailed morphological analysis of the embryonic development, as well as expression analysis and RNAi knockdowns of some of these genes point towards several strong similarities between the very complex *Thermonectus* principal eyes, and individual compound eye ommatidia. This raises the possibility that these functionally different eyes are derived from a compound eye ommatidium-like ancestor. One of the most exciting organizational features of *Thermonectus* principal eyes is the presence of bifocal lenses. To explore how they might be formed we extracted proteins from *Thermonectus* and *Drosophila* lenses and based on mass spectrometry identified 11 and 4 putative lens proteins, respectively. In both species, lens proteins are dominated by cuticular proteins, and in situ hybridization shows strong localization of some of the *Thermonectus* lens proteins to corneagenous cells that, like the lens-producing cone cells of *Drosophila*, extend to the base of the eye. Taken together, our study draws a unique connection between the development and evolution of functionally fundamentally different invertebrate eyes.

C10-03 Evolution of morphological complexity and modularity in the primate skull using anatomical network analysis

Esteve-Altava, Borja (University of Valencia, Paterna, ESP); Boughner, Julia (University of Saskatchewan, Saskatoon, SK, CAN); Diogo, Rui (Howard University, Washington DC, USA); **Rasskin-Gutman, Diego** (University of Valencia, Paterna, ESP)

Morphological complexity and modularity are two concepts closely related in evolution and development, whose study has traditionally taken different conceptual and methodological approaches. An integrative framework based on interaction among anatomical parts can unify both approaches: when interaction among parts forms morphological structures that develop and function semi-independently, morphological complexity arises in evolution due to modularity. However, most studies on modularity aim to identify a priori defined functional, genetic, developmental or variational boundaries in morphological structures, by analyzing patterns of correlated change in their size, shape, or growth rates during ontogeny or evolution. Anatomical Network Analysis (AnNA) has been specifically designed as an integrative tool to carry out morphological EvoDevo studies. Rather than testing a priori hypotheses about modularity, AnNA analyzes the structural relationships among parts, finding modules that can be further tested using other methods. The use of AnNA has proved fruitful to explore the evolution of morphological complexity concomitant to the reduction of the number of bones in tetrapods, the organization of modularity in the human skull, and their interplay in configuring the adult skull during ontogeny. Here we present our current research on the interaction among skull bones in Primates using AnNA, showing the relationship between morphological complexity and the modular organization of skull in a phylogenetic context. Our results allow us to understand, and formulate new questions about primate evolution and morphological variation in a broader morphological EvoDevo context.

C10-04 Elastin gene neo-functionalization endows teleost-specific heart component, "bulbus arteriosus", in fish development and evolution

Moriyama, Yuuta (University of Tokyo, JPN); Takeuchi, Jun K. (University of Tokyo, JPN); Koshiba-Takeuchi, Kauzko (University of Tokyo, JPN)

The vertebrate heart basically consists of four successive chambers; sinus venosus, atrium, ventricle and outflow tract (OFT). In teleost, OFT is called "bulbus arteriosus" (BA). It has been regarded as one of the most important evolutionary novelties in teleost evolution. BA has an elastic wall and functions as maintaining continuous blood flow into the gill arches regulating blood pressure wave. BA is a unique organ that is composed of smooth muscle like blood vessel while OFTs in other vertebrates including non-teleost fish are composed of cardiac muscle; teleost converts their cardiac OFT into BA composed of smooth muscle in their development and evolution. Through analyzing the morphogenesis of BA in zebrafish, we addressed the mechanism of cell fate determination into smooth muscle cells (SMCs) or cardiomyocytes (CM), which leads to the acquisition of evolutionary novelty. There are two elastin genes in teleost, *elastin1* and *elastin2*, compared with a single gene in other vertebrates. Elastin is the extracellular matrix protein (ECM) imparting the physiologically essential properties of extensibility and elastic recoil, and the major component of BA and blood vessel. We found that the expression pattern of *elastin2* in zebrafish embryos is restricted in BA, while *elastin1* is expressed not only in BA but also skeletal and nervous tissues. Furthermore, genetic architecture of *elastin2* is quite different from that of *elastin1*, which is similar (orthologous) to mammalian elastin gene. To investigate the function of elastin genes, we performed knockdown experiments and found that *elastin2* knockdown embryos exhibited hypoplasia of BA. Furthermore, we found that the ectopic cardiac muscle in BA of *elastin2* knockdown embryos. These results imply that the teleost-specific ECM *elastin2* positively regulates cell fate determination into SMCs not into CM, and contributes to acquisition of BA in teleost development and evolution.

17.20 – 18.00 Keynote Lecture (K2)

A "Devo-Evo" approach to unravel the hidden logic of cell and tissue polarity in plants and animals

ROOMS C1&2

Veronica Grieneisen
(John Innes Centre, Norwich, GBR)
Chair: Charlie Scutt

In this talk I wish to compare and contrast cell and tissue polarity between very diverse organisms, with the core focus on how conserved elements on the cellular level — together with different evolutionary constraints on the level of multicellularity — can provide a unifying framework and novel view on planar cell polarity in animals, while also consolidating divergent models ("up-the-gradient" and "with-the-flux") of hormone-patterning mechanisms in plants. This framework allows us to understand how major developmental processes in plant development can be modulated and controlled through basic mechanism of cell polarity. Computational approaches combined with molecular studies and *in vivo* microscopy were necessary to reveal how polarity is coordinated and linked on three different levels: on the scale of the tissue, the cellular and subcellular tissue level. At the single cell level, a spatially uniform activation and patterning of GTPases can cause polarity to emerge spontaneously, independently of spatial pre-patterns or localized polarizing signals. We argue that plants and animals have inherited this same "unicellular mode" of establishing

cell polarity, and that multicellular coordination has thereafter diverged using this underlying mechanism as a building block: Being capable of intracellular partitioning, neighboring plant cells that are separated by cell wall then coordinate their polarities through indirect cell-cell coupling. This is resultant from changes in concentration level of a phytohormone, auxin, along cells. In the specific case of pavement cells of leaves (jigsaw-shaped cells with interlocking lobes and indentations), this phenomenon comes about as interdigitation, and requires the opposite response of identical neighboring cells to the same local auxin signal in the cell wall, between the cells. Our theoretical work identifies key requirements for such indirect cell-cell signaling that that gives rise to correct interdigitation. These requirements, based on known molecular interactions, can then be extrapolated to other multi-cellular tissues to understand the interdependency between cell and tissue polarity. Extrapolating these findings we further show how animal cells, capable of direct cell-cell coupling, can establish, through similar principles, robust tissue coordination.

18.00 – 20.00 Poster Session 1
Corridors C and ROOM E
(even numbers)

20.30 Reception at Vienna City Hall

Thursday, July 24th

09.00 – 10.40 Symposium S9:
Plant EvoDevo: Linking cross-species genetic and morphological variation

ROOM A

Organizers: John Bowman and Christian Hardtke
Chair: Christian Hardtke

S9-01 **Inverted regulatory logic in hormone pathway interactions shapes different root system types**
Hardtke, Christian (University of Lausanne, CHE)

The activity of various plant hormones, notably auxin, is pivotal for root system development in tracheophytes. For instance, local auxin biosynthesis is essential for root formation and growth in the dicotyledon model, *Arabidopsis thaliana*. Thus, increasing interference with auxin biosynthesis results in increasingly less branched and shorter roots, partly because of reduced cell elongation. However, counterintuitively, mutants in an auxin biosynthesis pathway enzyme in the monocotyledon model, *Brachypodium distachyon* display more branched and dramatically longer seminal roots, because mature cells are thinner, more elongated and therefore more anisotropic than in wild type. Interestingly, this phenotype can be mimicked in wild type by pharmacological interference with production of a key auxin biosynthesis intermediate, but also by interference with the biosynthesis or signalling pathway of another plant hormone, ethylene. The latter positively controls auxin biosynthesis in *Arabidopsis* roots. Surprisingly however, auxin levels in the *Brachypodium* mutants are elevated rather than reduced, because of a simultaneous up-regulation of the second, rate-limiting step of the pathway. Ethylene normally represses this second step, suggesting an inverted regulatory relation between the two hormones in *Brachypodium* as compared to *Arabidopsis*. Our results point to a complex homeostatic crosstalk between auxin and ethylene in *Brachypodium* roots, which is fundamentally different from *Arabidopsis* and could in part be responsible for the divergent root system morphologies of these two model plants. This inverted regulatory logic might be conserved in other monocotyledons, such as rice, and could play a role in the developmental response to flooding.

S9-02 Genetic determinants of petal number variation between *Arabidopsis thaliana* and *Cardamine hirsuta*

Monniaux, Marie (Max Planck Institute for Plant Breeding Research, Cologne, GER)

Understanding the genetic basis of morphological evolution is a key question in biology, and bridging the gap between genotype and the final growth and shape of an organ is still challenging. Here, we use two closely related Brassicaceae species: *Arabidopsis thaliana* and *Cardamine hirsuta*, to identify the genetic determinants of variation in their petal number. While *A. thaliana* flowers always exhibit 4 petals, *C. hirsuta* flowers have a variable number of petals, ranging from 0 to 4. The development of *C. hirsuta* as a model genetic system means that this comparison with *A. thaliana* constitutes an ideal case study to understand general mechanisms of morphological evolution. We identified the A-class MADS-box gene *APETALA1 (AP1)* as a key determinant of petal number in both species, and transferring the AP1 genomic locus of *A. thaliana (AtAP1)* into *C. hirsuta* plants is sufficient to restore the ancestral crucifer petal number of four. Subtle differences in the expression patterns of *AtAP1* and *ChAP1* in the flower meristem, at the stage relevant for petal initiation, suggest that regulatory differences at the *AP1* locus may underlie petal number variation between these species. We aim to identify the *cis*-regulatory changes between *AtAP1* and *ChAP1* promoters that are responsible for their specific expression and function, and determine the target genes of both regulators and their effect on organ growth and geometry.

S9-03 A division in PIN-mediated Auxin patterning during organ initiation in grasses

O'Connor, Devin (Cambridge University, GBR); Runions, Adam (University of Calgary, AB, CAN); Sluis, Aaron (University of California Albany, CA, USA); Bragg, Jennifer (USDA, Albany, CA, USA); Vogel, John (USDA, Albany, CA, USA); Prusinkiewicz, Przemyslaw (University of Calgary, AB, CAN); Hake, Sarah (University of California, Albany, CA, USA)

The hormone auxin plays a crucial role in plant morphogenesis. In the shoot apical meristem, the PIN-FORMED1 (PIN1) efflux carrier concentrates auxin into local maxima in the epidermis, which position incipient leaf or floral primordia. From these maxima, PIN1 transports auxin into internal tissues along emergent paths that pattern leaf and stem vasculature. In *Arabidopsis thaliana*, these functions are attributed to a single PIN1 protein. Using phylogenetic and gene synteny analysis we identified an angiosperm PIN clade sister to PIN1, here termed *Sister-of-PIN1 (SoPIN1)*, which is present in all sampled angiosperms except for Brassicaceae, including *Arabidopsis*. Additionally, we identified a conserved duplication of *PIN1* in the grasses: *PIN1a* and *PIN1b*. In Brachypodium distachyon, *SoPIN1* is highly expressed in the epidermis and is consistently polarized toward regions of high expression of the DR5 auxin-signaling reporter, which suggests that *SoPIN1* functions in the localization of new primordia. In contrast, *PIN1a* and *PIN1b* are highly expressed in internal tissues, suggesting a role in vascular patterning. *PIN1b* is expressed in broad regions spanning the space between new primordia and previously formed vasculature, suggesting a role in connecting new organs to auxin sinks in the older tissues. Within these regions, *PIN1a* forms narrow canals that likely pattern future veins. Using a computer model, we reproduced the observed spatio-temporal expression and localization patterns of these proteins by assuming that SoPIN1 is polarized up the auxin gradient, and *PIN1a* and *PIN1b* are polarized to different degrees with the auxin flux. Our results suggest that examination and modeling of PIN dynamics in plants outside of Brassicaceae will offer insights into auxin-driven patterning obscured by the loss of the *SoPIN1* clade in Brassicaceae.

S9-04 Towards understanding the genetic basis for diversification of leaf forms

Tsiantis, Miltos (Max Planck Institute for Plant Breeding Research, Cologne, GER)

A key challenge in biology is to understand how diversity in organismal form is generated. Genetic analyses in model systems have identified key regulators that sculpt the body plans of metazoa and seed plants. However, less is known about how the action of such regulators produces particular organ shapes, or how the balance of conservation versus divergence of such form regulating pathways generated the tremendous morphological diversity of multicellular eukaryotes. One impediment to answering these questions is the relative paucity of experimental platforms where genetic tools can be utilized to unambiguously study morphogenesis and its evolution in a genome-wide, unbiased fashion. To circumvent this problem we developed the *Arabidopsis thaliana* relative *Cardamine hirsuta* into a versatile system for studying morphological evolution. We aim to understand the molecular mechanisms through which leaf morphology evolved in these species, resulting in simple, undivided leaves in *A. thaliana* and dissected leaves with distinct leaflets in *C. hirsuta*. This presentation will discuss our progress towards understanding the genetic pathways that specify dissected versus entire leaf shapes and that regulate the number, position and timing of leaflet production.

09.00 – 10.40 **Symposium S10:**
What should bioinformatics do for EvoDevo?

ROOM B

Organizers: Günter Plickert, Mark Blaxter, Paula Mabee and Ann Burke
Chair: Paula Mabee

S10-01 Bioinformatics for Evo Devo: Connecting evolutionary morphology and model organism genetics

Mabee, Paula (University of South Dakota, Vermillion, SD, USA)

The Phenoscape team has developed a systematic bioinformatics method for proposing candidate genes for evolutionary novelties. That is, using a specific phenotypic feature as an entry point, an evo-devo researcher can immediately retrieve all of the genes from model organisms that are known to have an effect on that phenotype. This is the core element of the evo-devo approach, but how can it be extended to accommodate the additional needs of the community? The Phenoscape system has prototyped its approach using genetic data from the vertebrate model organisms (zebrafish, Xenopus, mouse, human) and evolutionary data from the phylogenetic literature, with a current focus on the fin/limb transition. Ontology-based methods are used to represent the uncomputable phenotypic features in free-text format from the comparative literature. This enables automatic linkage to phenotypes resulting from genetic manipulations. We have tested two *in silico* candidate gene proposals for evolutionary novelties in the 0wet-lab. Specifically, we selected two features that characterize Siluriformes (catfishes), i.e., the loss of scales and absence of basihyal, and we examined the computer predicted tissue-specific expression patterns of candidates eda, edar, and brpf1 in the channel catfish, *Ictalurus punctatus*. As predicted, brpf1 and eda were not expressed in the appropriate tissues at the expected developmental stage. These data support the broad-scale utility of this approach to integrate genetic and phenotypic data in formulating devo-evo hypotheses. Morphological data in this computable format can be browsed, sorted and aggregated in ways that present unprecedented possibilities for data mining and discovery. Current work by the team includes development of methods for a "phenoblast" that accommodates multiple phenotypes. Additionally, a tool for aggregating phenotypic data from multiple studies into a matrix that can be viewed on phylogenetic trees has recently been developed. The reasoning enabled by the informatics approach allows inference of presence/absence data across great taxonomic breadth. We demonstrate this by using examples from fin and limb data in tetrapod evolution and fin presence/absence data in actinopterygian fishes.

S10-02 Insights into the evolution and development of planarian regeneration from the genome of the flatworm, *Girardia tigrina*

Kumar, Sujai (University of Oxford, GBR); Kao, Damian (University of Oxford, GBR); Aboobaker, Aziz (University of Oxford, GBR)

Next-generation sequencing technologies have dramatically transformed the study of non-model organisms that can provide new insights into evolutionary developmental biology. We have sequenced, assembled, and annotated the genome of *Girardia tigrina*, a planarian that can perform whole-body regeneration (WBR) complete with a new head, eyes, and neurons, even from a small fragment of its tail. We show examples of how the genome has not only assisted traditional genetics experiments, but has also allowed us to use comparative genomics approaches to identify new candidates for examining the molecular bases of WBR. Although sequencing this relatively large genome (1.5 Gbp) using only short-read technologies was inexpensive, there were many challenges that had to be overcome. We demonstrate how we solved these problems using innovative bioinformatics pipelines. Overall, bioinformatics can help create genomic and transcriptomic resources for many more such non-model species as sequencing costs drop. Armed with multiple genomes for many species with a range of regenerative abilities, we will be able to better understand the mechanisms underpinning WBR.

S10-03 From the wet lab to the computer and back: A stage specific RNAseq analysis elucidates the molecular underpinnings and evolution of Hydrozoan development

Schiffer, Philipp (University of Cologne, GER)

For a long time, wet-lab biological research and bioinformatics have been separate disciplines distinguished by a great divide of understanding. On the one side was the search for function in single genes, on the other the hunt for signal and context in massive amounts of data. Also, to conduct bioinformatics analyses large collaborations were necessary to ensure the funding needed for sequencing genomes or ESTs. However, with the advent of 2nd generation sequencing large scale analyses have become feasible to single laboratories. Still, organism-based laboratories often lack the bioinformatic facilities and background to analyse the data, while bionformaticians often lack the handle to appreciate classical wet lab work with real life forms. Clearly an exchange between both methodologies is needed. In this talk I will illustrate how a lab-trained, turned computational biologist got involved into the analysis of the fascinating biology of an early branching metazoan species, the hydrozoan model *Hydractinia echinata*. This will serve as an example of how the interplay between

computational analysis and wet lab experiments can help contribute to identify conserved traits in the evolution of developmental control outside of bilateria. To investigate gene expression patterns RNAseq is the 2nd generation sequencing method of choice. We used an RNAseq based approach to analyse several life-cycle stages of *H. echinata*. We combined a developmental time course experiment to unravel stage-specific expression differences during embryomic development with the analyses of maternal deposition in comparison to zygotic expression. To elucidate the molecular background of differentiation between sexes in metazoa, we also compared transcriptomes of male and female *Hydractinia*. For further downstream analyses we have now established a workflow where annotated candidate genes from our broad computer based differential expression inferences are first functionally characterised on a gene-by-gene level and then re-confirmed in the RNAseq data. Next individual candidate genes can be analysed by reverse genetics available in *H. echinata* and then put into the broader context of our knowledge on evolution of developmental control mechanisms. Our data will shed light on the evolution of developmental processes such as gastrulation, deployment of light perception or myogenesis in Cnidaria as well as in comparison to Bilateria.

S10-04 Insights into the evolution of early development of parthenogenetic nematodes by second generation sequencing

Kraus, Christopher (University of Cologne, GER); Schiffer, Philipp (Universität of Cologne, GER); Kroiher, Michael (University of Cologne, GER); Schierenberg, Einhard (University of Cologne, GER)

In many sexually reproducing metazoans the sperm entry point is an important determinant of the first cleavage. It induces polarity establishment and axis formation in invertebrates and vertebrates. However, in nearly every metazoan phylum parthenogenetic species exist and in most cases they lack sperm. This prompts the question: How is polarity establishment regulated in species without sperm on a molecular level? In a first attempt to investigate this scientific question, we chose the diverse phylum of nematodes and selected closely related species with sexual and asexual reproduction. In addition, we used the genetic model *Caenorhabditis elegans*, for which the gene regulatory network of polarity establishment in the zygote is functionally elucidated. Unfortunately, for most other nematodes neither forward nor reverse genetic techniques are feasible. Hence, we chose an alternative approach that consisted of (1) genomic sequencing, (2) RNAseq of IVT amplified mRNA, (3) orthology screening, and (4) bioinformatic data mining for de-novo functional annotation of second generation sequencing data. This approach gave us access to the genetic background of the selected nematodes and more specifically we could focus on early development, by stage-specific sampling of embryos during the first cleavages. The orthology screening allowed direct comparisons of orthologs among the selected nematodes. Combining these lab-based and bioinformatic approaches, we were able to determine molecular differences between the selected species. We found most differences between the *Caenorhabditis* genus and other nematodes. Most strikingly, our orthology screening showed that regulators of early polarity establishment such as PAR-2, LIN-5 and GPR-1/-2 are missing in nematodes outside the *Caenorhabditis* genus. In contrast to that our RNAseq and genomic sequencing data indicated candidate genes potentially involved in polarity establishment. We conjecture that the genetic background of nematodes outside the *Caenorhabditis* genus is a prerequisite for the evolution of parthenogenetic species in these taxa.

09.00 – 10.40 Symposium S11: Ecological and environmental impacts on the evolution of organismal development I

ROOM C1

Organizers: Chris Lowe, John Willis and Angelika Stollewerk
Chair: Angela Stollewerk

eden Evo-Devo-Eco Network

S11-01 EcoEvoDevo and the origins of morphological complexity in the worker caste in ants

Rajakumar, Rajendhran (McGill University, Montreal, QC, CAN); Fave, Marie-Julie (McGill University, Montreal, QC, CAN); **Abouheif, Ehab** (McGill University, Montreal, QC, CAN)

Integrating evo-devo into the mainstream of evolutionary theory requires an understanding of the complex and non-linear interactions between environment, organismal development, and evolution. Ants provide a powerful system to understand these interactions because they are social and polyphenic, which means that queens and workers within the colony are determined by environmental, social, and ecological interactions during development. As a consequence, ants are one of the most morphological diverse, ecologically dominant, and evolutionarily successful insects on our planet. I will present evidence to show that although workers are completely wingless, vestigial wing development in ants integrates environmental and social signals

to control worker caste regulation, differentiation, and evolution. These results have important implications for understanding how environmental and ecology impacts levels of selection, co-option, developmental system drift, and evolution of novelty and complexity.

S11-02 Inducible defenses in Daphnia

Laforsch, Christian (University of Bayreuth, GER)

Predation is a key factor in the evolution of prey species and the dynamics of prey communities. In both animals and plants, different defensive mechanisms have evolved in response to this selection pressure. The term phenotypic plasticity describes the ability of a genotype to produce different phenotypes in response to different environmental conditions. Phenotypic plasticity in defensive traits appears to be an appropriate mechanism to cope with the variable hazard of a frequently changing predator spectrum. Phenotypic plastic responses enable prey organisms to express a particular defence only if a reliable cue for a future attack is present. Thereby, the organisms can minimise costs affiliated with the formation of a defence when predation risk is low. A single genotype can change its life history, behaviour and morphology as an adaptation to predation risk. Formations of protective devices in Daphnia (Crustacea), such as helmets and spines, are prominent examples of these chemically induced defenses in response to predator stress. To uncover the mechanisms of phenotypic plasticity has been always a main goal in evolutionary ecology. Combining proteomics and genomics provide an exceptional opportunity to reveal the nature and complexity of plastic defensive traits in Daphnia on a molecular basis.

S11-03 Genetics of larval mode in the poecilogonous polychaete, *Streblospio benedicti*

Rockman, Matthew (New York University, NY, USA)

Among marine animal lineages, evolutionary transitions between planktotrophic larval forms with long dispersal periods and lecithotrophic larval forms that can settle without feeding or dispersing are ubiquitous. These transitions modify morphology, physiology, life history, and population biology. The polychaete *Streblospio benedicti* provides a unique model for transitions in larval form, as this species is poecilogonous, producing both planktotrophic and lecithotrophic larvae. Larval form in *S. benedicti* is highly heritable, with little influence of environmental factors. We describe genetic analysis of larval form in *S. benedicti*, demonstrating that distinct aspects of larval form are genetically separable and are shaped differentially by maternal-effect and zygotic variation. We further demonstrate via population-genomic analyses that most of the genome in these animals experiences extensive gene flow independent of larval type, while a small number of genomic regions is highly differentiated. Discovery of the genetic mechanisms underlying this life-history polymorphism will allow dissection of the ecological factors that maintain it in nature and drive larval evolution among lineages.

S11-04 Determination of sexual fate in the temperature dependent red-eared slider turtle, *Trachemys scripta elegans*

Capel, Blanche (Duke University, Durham, NC, USA); Czerwinski, Mike (Duke University, Durham, NC, USA); Mork, Lindsey (Duke University, Durham, NC, USA); Looger, Loren (Janelia Farm Research Campus, Ashburn, VA, USA); Natarajan, Anirudh (Duke University, Durham, NC, USA)

In mammals, sex determination is controlled by antagonistic signaling pathways that are activated by expression of the male sex-determining gene on the Y-chromosome, Sry. In contrast to this strong genetic mechanism, the incubation temperature of the egg determines sexual fate in many reptilian species, including the red-eared slider turtle, *Trachemys scripta elegans*. In this species, embryos incubated at low temperatures during the initial stages of gonad formation develop as males, while those kept at higher temperatures develop as females. Whether cryptic genetic mechanisms exist in this temperature dependent species, and whether underlying pathways are similar to those in mammals is unknown. In *T. scripta*, incubation at the threshold, or pivotal, temperature (PvT) results in an even ratio of males and females, and rarely produces an intersex individual. One possibility is that sexual fate is stochastic at the PvT, but coordinated by systemic signals within a single embryo. To test this possibility, we explanted gonads from individual embryos to culture and showed that gonad pairs incubated separately at the PvT share a strong predisposition for one sex or the other when cultured in isolation. Our findings suggest that the outcome of sex determination in these reptiles is heavily influenced by an inherent predisposition at the PvT that could be genetic or could be established by earlier influences in the embryo. To identify factors regulating sex determination at male- and female-producing temperatures, we sequenced the gonad transcriptome of the red-eared slider turtle. In the absence of an assembled genome, we developed a de novo transcript assembly pipeline and homolog based annotation from gonad and non-gonadal tissue samples at the male and female producing temperatures. We generated a transcript database for the assessment of gene and transcript level differential expression, and identified a comprehensive list of differentially expressed genes, including transcription factors and signaling pathways that underlie sex determination in *T. scripta*.

09.00 – 10.40 Symposium S12: Developmental basis of quantitative variation

ROOM C2

Organizers: Mihaela Pavlicev and Günter Wagner
Chair: Mihaela Pavlicev

S12-01 The evolution of pleiotropy in relation to integration, modularity, and individuation

Cheverud, James M. (Loyola University, Chicago, USA)

The principle of modularity plays an important role in our understanding of morphological evolution. Modules are sets of integrated traits that are semi-independent from other such sets. Examples include the mammalian fore- and hindlimb, vertebrate vertebral and dental morphology, and mammalian digital morphology. Modularity can both facilitate and constrain adaptive responses to selection depending on the modular structure in relation to selection. When a coordinated adaptation of traits is selected, modularity facilitates adaptation by limiting the potentially negative consequences of correlated responses of features in different modules. However, if selection is on only a subset of traits in a module, their integration can constrain evolution. Given a baseline of a fully integrated set of features, the evolutionary mechanism by which such features can be parcellated or individuated remains unclear. With quantitative trait locus mapping, we show that genetic variation in pleiotropy is abundant for morphological traits in mice being produced by different patterns of epistasis for different traits. These loci represent genotypic differences in the relationships between traits without necessarily affecting trait means. We further show that directional, adaptive selection will enhance genetic variation in the selected dimension, while limiting it in all other dimensions, sculpting new modules configured to the new selective environment. This process may be responsible for making and breaking modules thereby limiting the evolutionary constraint caused by integration while facilitating the separate adaptation of parts.

S12-02 Mutational variation in epistatic pleiotropy and the genotype-phenotype map of multi-drug resistance in HIV-1

Guillaume, Frederic (University of Zurich, CHE)

Pleiotropy and epistasis are key features of genetic systems and both play a fundamental role in the evolutionary process of adaptation. Nevertheless, the evolutionary significance of epistasis, the effect of gene interactions, is still unclear and debated. By contrast, pleiotropy, whereby a gene affects multiple phenotypes, is known to strongly affect the adaptive capacity of a population by shaping the distribution of genetic co-variation among phenotypic traits and cause evolutionary constraints. I will show that pairwise epistasis may significantly shape the pattern of genetic co-variance among fitness-related traits in the HIV-1 virus by modifying the pleiotropy of single mutations in a set of 1,859 amino-acid substitutions in the reverse transcriptase and protease genes of HIV-1. Epistatic pleiotropy, defined as the non-additive effect of interactions on the pleiotropic degree of double mutations, determines the modular structure of the in-vitro replicative capacity of the virus in 15 drug environments. This analysis shows that epistasis is central to the structure of the genotype-phenotype map of drug resistance in HIV-1 and to its adaptive capacity.

S12-03 Highly monotone genotype-phenotype maps emerging from gene regulatory networks

Gjuvsland, Arne (Norwegian University of Life Sciences, Ås, NOR); Wang, Yunpeng (Norwegian University of Life Sciences, Ås, NOR); Plahte, Erik (Norwegian University of Life Sciences, Ås, NOR); Omholt, Stig W. (Norwegian University of Science and Technology, Trondheim, NOR)

In quantitative genetics population-level phenomena, such as heritability are explained in terms of properties (e.g., additivity, dominance, epistasis) of the genotype-phenotype (GP) map. This level of explanation involves measuring gene action in phenotypic units, whereas a future theory should also offer explanations based on how genes actually work and interact in complex networks. Towards this end we study gene regulatory network models where genetic variation is introduced in process parameters while expression levels are used as phenotypes. We have earlier examined the effect of feedback on populations-level components of genetic variance and shown that regulatory networks with positive feedback gives rise to more statistical epistasis than those with negative or no feedback. Motivated by these clear signatures we seek to understand the types of constraints that the regulatory architecture of biological systems put on the GP map. We hypothesize that design principles of regulatory systems lead to largely monotone gene action, i.e., order-preservation between allele content and corresponding genotypic values in the mapping from genotypes to phenotypes. We develop measures of monotonicity in genotype-phenotype maps and show that in random GP maps monotonicity and additivity are strongly correlated. Next we perform a simulation study of functional genetic variation in 1881 different three-gene regulatory networks and study the emergent genotype-phenotype maps. In general the resulting GP maps are highly monotonic across network types, explaining how highly nonlinear biological systems can give rise to mainly additive genetic variance. However, we also reveal clear differences between classes of networks, in particular networks involving incoherent feedforward and positive feedback promote non-monotone gene action. Furthermore

we derive analytical results describing how genetic variation in a locus is propagated through a network depending on regulatory chains and loops and use it to generalize our simulation results to larger networks. Our work is a step towards a theory capable of explaining the pleiotropy and epistasis features of genetic variation in complex regulatory networks as functions of regulatory anatomy and functional location of the genetic variation.

S12-04 Managing the pleiotropy of "pleiotropic" transcription factor genes

Wagner, Günter (Yale University, New Haven, CT, USA); Lynch, Vincent (The University of Chicago, IL, USA); Nnamani, Mauris (Yale University, New Haven, CT, USA); Pavlicev, Mihaela (University of Cincinnati, OH, USA)

One of the arguments against the plausibility of transcription factor evolution is that most transcription factor genes have roles in many different developmental and physiological contexts, i.e. are pleiotropic. The reasoning is that any amino acid substitution in a pleiotropic transcription factor will affect a number of characters and thus likely have at least one if not many deleterious effects. On the other hand there is plenty statistical evidence that transcription factor genes undergo adaptive evolutionary change and thus must be able to limit the number of pleiotropic effects a mutation has. In our studies of transcription factor evolution we asked what the molecular mechanisms are that limit the pleiotropic effects of amino acid substitutions in a "pleiotropic" transcription factor. In this contribution we will discuss our research on HoxA11, a transcription factor with roles in limb development, male and female reproductive system, cloaca development, kidney development and others more. A recent derived role of HoxA11 is its role in mammalian pregnancy, specifically in decidual cells of the uterine endometrium. We present evidence that the derived transcription factor activity depends on the presence of another transcription factor, Foxo1a. The mechanism for the derived function is one where the derived HoxA11 protein is an activator in the presence of Foxo1a and S/T phosphorylation. Based on these results we suggest that character specific effects of pleiotropic transcription factors are possible through character specific interactions with other co-expressed transcription factors.

11.10 – 12.50

Symposium S13: Structural organization in vertebrate dentitions: Molecules, morphology and function

ROOM A

Organizers: Abigail Tucker and Moya Smith
Chair: Moya Smith

S13-01 Teeth inside and outside the mouth: Topographic relationships in sawshark and sawfish dentitions (Elasmobranchii; Chondrichthyes)

Welten, Monique (Natural History Museum, London, GBR); Meredith Smith, Moya (King's College London, GBR); Underwood, Charlie (University of London, GBR); Fraser, Gareth (University of Sheffield, GBR); Johanson, Zerina (Natural History Museum, London, GBR)

Sharks and rays have been studied extensively to address the origin and evolution of teeth in jawed vertebrates. They possess a micromeric dermal skeleton in which odontodes form as separate placoid scales (skin denticles) in a pattern distinct from teeth on the jaws. Extended rostra are present in taxa of both sharks and rays represented by the sawsharks (*Pristiophorus*, *Pliotrema*), sawfish (*Pristis, Anoxypristis*) and the fossil Sclerorhynchoidea. In these taxa, tooth-like structures on the rostra differ from both oral teeth and placoid scales, presenting a challenge in understanding the morphogenesis of this rostrum pattern, and whether it is related to that of the dentition. Sawshark, sawfish and sclerorhynchid rostra are convergently evolved feeding and/or sensorial structures. Sawfish retain the same set of rostral "teeth" during their whole life, each growing from the base via the deposition of dentine. These "teeth" are of equal size and equal spacing, located in deep sockets on the lateral side of the rostrum. In contrast, all "teeth" in sawsharks are replaced. Sawshark rostral "teeth" are of different sizes and include three distinct topographic series; lateral "teeth" (lateral rostral denticles) of varying sizes, located in shallow pits on the calcified cartilage of the rostrum; a second series (lateral cephalic denticles) extends from the lateral "tooth" row caudally towards the mouth, while a third series (ventral rostral denticles) is present in a different pattern, extending rostrally from the nasal capsules. It is notable that sclerorhynchids present the same morphological pattern; these rostral "teeth" are replaced in both groups. X-mCT provided rendered volume densities of 3D models that were used to study growth, replacement and patterning of "teeth", including segmentation and measurements of functional and unerupted "teeth". For example, in the sawshark, new additions to lateral rostral "teeth" and their patterning appear to be the result of growth rostrally, while replacement "teeth" occur irregularly, only after "tooth" loss. Although irregular, this replacement is in a size-specific manner, and in size-dependent locations on the rostrum. Their

orientation, size order and replacement suggest axial patterning and regulation. Our hypothesis is that these "teeth" represent neomorphic dermal structures on the rostrum, patterned independently of oral teeth and evolved separately. A mineralized cartilaginous rostrum, with these lateral (and ventral) denticles occurs only in chondrichthyans, but appears to have evolved independently on at least three separate occasions within the group.

S13-02 Mechanical constraints during cusp pattern development in rodent molars

Renvoise, Elodie (University of Helsinki, FIN); Kavanagh, Kathryn (University of Massachusetts, Dartmouth, MA, USA); Kallonen, Aki (University of Helsinki, FIN); Häkkinen, Teemu (University of Helsinki, FIN); Rice, Ritva (University of Helsinki, FIN); Jernvall, Jukka (University of Helsinki, FIN)

The tooth crown pattern, and especially the crown complexity, was shown to be functionally related to the animal diet. However, the crown morphogenesis is only determined during embryonic stages. Developmental biology and computational modeling have shown that the cusp pattern of teeth can be self-regulated by signaling molecules associated with physical properties of the cells, independently from the surrounding tissues. This idea of self-regulation of the crown pattern formation is reinforced by the fact that molecular perturbations in the tooth system can reproduce crown phenotypes that appeared during evolution. During evolution, the arvicolines (voles, lemmings and muskrats) have developed an extreme alternated cusp pattern in their molar crown that never happened as such during rodent evolution. In 2002, Salazar-Ciudad and Jernvall introduced a lateral bias parameter in their morphodynamic model of molar development. Together with anterior elongation, these biases influenced whether cusp patterns were alternate or confluent. The nature of these biases has not been tested experimentally, and here we examined for the first time the possible role of the jawbone as a mechanical constraint in fine-tuning of cusp patterns. The cusp offset of the first lower molar (m1) in vole is lost when the tooth is cultured out from the jaw. Laterally constrained mouse and vole m1 *in vitro* recovers the vole-like cusp pattern offset. In voles, the thickness of the jawbone is larger than in mouse and shows a bucco-lingual asymmetry. 3D reconstructions of m1 and jawbone development *in vivo* showed a constraint of the jawbone in vole, while the m1 of mouse grows better laterally, "pushing" aside the jawbone. Therefore, the bucco-lingual constrain of the jawbone during the *in vivo* development can generate the alternate cusp pattern of voles. In conclusion, we show how the cusp pattern morphogenesis in molars can be influenced by a lateral constraint of the jawbone. This lateral constraint of the jawbone, may explain the variability of the crown patterns of molars in the mosaic evolution of rodents.

S13-03 Development and fate of the dental lamina in amniotes

Buchtova, Marcela (Academy of Sciences of the Czech Republic, Brno, CZE)

The dental lamina arises as an epithelial thickening in the oral epithelium. Later, the lamina grows deeply into the mesenchyme and teeth germs bud in specific directions from the lamina. It seems to be a correlation between dental lamina morphology and the number of replacement dental generations in Amniotes. Some reptilian species with constant teeth replacement exhibits large dental lamina. Furthermore, reptilian lamina is continuous along the jaw and remains as a compact structure connecting the individual generations of teeth during pre-hatching and post-hatching stages. The successional dental lamina is formed after the initiation of the first teeth generations. The timing of successional lamina appearance is species-specific and generally arises earlier in the dentition with simple tooth shapes. In diphyodont mammalian species, the dental lamina disintegrates early in embryogenesis into small epithelial clusters, which disappear by similar mechanism to the degradation of palatal seam during secondary palate closure. The l amina loses the basal membrane that allows the epithelial cells to break away from the lamina and migrate into the surrounding mesenchyme. In a monophyodont mammals, only short connection between the tooth and the oral epithelium develops (also called the dental stalk). Tooth anlagen stays in contact with the oral epithelium through tooth development up to its eruption during postnatal stages. However, bays of mesenchymal cells with numerous capillaries disrupt the integrity of epithelial tissues. Despite of this fact, ultrastructural analysis revealed that epithelial cells are connected together by large desmosomes and their connections stay functional up to the teeth eruption. Numerous degenerated mitochondriae and long bundles of cytoplasmatic filaments fill the epithelial cells before teeth eruption and cornification of the lamina occurs in some areas with keratohyaline granules accumulation similar as in the oral epithelium. These groups of lamina cells become a part of gingiva while areas above the cusps undergo an apoptosis and they are peeled off from the surface. Furthermore, small successional lamina is initiated in a mouse on the lingual side of the tooth germ at late embryonic stages. However, it regresses during postnatal stages and replacement tooth formation is never initiated. In conclusion, as the fate of dental lamina cells is species-specific and an epithelial residue of dental lamina can form cysts and ameloblastomas, the understanding of developmental processes contributing to their persistence or disappearance became a key issue.

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S13-04 What the Myotragus evolutionary lineage tell us about the selective pressures that drive dentition patterning in mammals

Jordana, Xavier (Institut Català de Paleontologia Miquel Crusafont, Bellaterra, ESP); Moncunill-Solé, Blanca (Institut Català de Paleontologia Miquel Crusafont, Bellaterra, ESP); Köhler, Meike (Institut Català de Paleontologia Miquel Crusafont and ICREA, Bellaterra, ESP)

Because insular ecosystems are much simpler than mainland ones, islands are regarded as "natural laboratories of evolution". Insular ecosystems are intrinsically resource limited because their restricted landmass can only support a narrow number of primary producers, which in turn affects the energy flow at higher trophic levels. Therefore, as a rule, island ecosystems are impoverished of interspecific competitors and of predators, conditions that force a shift from a density-independent (r-selection) to a density-dependent (K-selection) selective regime. In an ecological context of high intraspecific competition for scarce resources and of low predation pressure, selection favours those traits related to more efficient energy intake and energy use. The fossil bovid *Myotragus* Bate, 1909 (Artiodactyla, Caprinae) from Mallorca island displays a series of derived phenotypic traits including dwarfing, reduced size of brain and sense organs, and frontalized orbits, among others, as a result of 5 million years of evolution under the unique ecological conditions of insular ecosystems. Some of those derived traits are shared by a large number of insular endemics, a phenomenon coined the "island syndrome". But the most striking modification in Myotragus is related to the dentition, which changes gradually during the anagenetic evolution of the lineage. The late Pleistocene and Holocene *M. balearicus*, the terminal species of the extinct genus, displays a dentition patterning unique among extant ruminants. *M. balearicus* presents one rodent-like evergrowing incisor en each hemimandible, a very reduced number of premolars and highly hypsodont molars (the dental formula for an adult is I 0/1, C 0/0, P 2/1, M 3/3). Their eruption sequence is also unusual among living bovids, as the permanent incisors and the premolars erupt relatively early. Our research on Myotragus suggests that their unique dentition patterning evolved as a functional adaptation to promote increased feeding efficiency and increased durability of dentition over a longer lifespan in response to resource limitation. Myotragus, hence, shows us that significant changes in dentition patterning may evolve in a scenario of density-dependence where changes in environmental conditions or shifts in feeding habits play only a secondary role.

11.10 – 12.50 Symposium S14: What should bioinformatics do for EvoDevo? II

ROOM B

Organizers: Günter Plickert, Mark Blaxter, Paula Mabee, and Ann Burke
Chairs: Günter Plickert and Mark Blaxter

S14-01 Petaloidy, polarity and pollination: The evolution of organ morphology networks

Specht, Chelsea (University of California Berkeley, CA, USA); Yockteng, Roxana (University of California Berkeley, CA, USA); Almeida, Ana M. R. (University of California Berkeley, CA, USA); Pineyro-Nelson, Alma (University of California Berkeley, CA, USA)

For the past decades, studies in flower EvoDevo relied heavily on a candidate gene approach, investigating correlations between expression patterns of candidate genes described from model systems with observed morphological diversification across lineages of flowering plants. While many of these candidate genes are conserved across vast evolutionary time, differences in copy number, expression patterns, and functional motifs make it increasingly challenging to assign homology of function to genes described from model systems. With whole genome and organ-specific transcriptome data, we are increasingly able to develop hypotheses for functional gene networks that control specific aspects of floral development, and investigate the evolution of these gene networks and their impact on the evolution and diversification of floral form. Here we present how a shift from thinking about "organ identity" to focusing on the processes that underly organ morphology can enable us to ask relevant questions and test relevant hypotheses about the evolution of floral organ morphology in diverse lineages of flowering plants.

S14-02 Aligning phonemes and genomes to understand the evolution of multicellular organisms

Donoghue, Philip (University of Bristol, GBR); Deline, Bradley (University of West Georgia, Carrollton, GA, USA); Greenwood, Jennifer (University of Bristol, GBR); Taylor, Richard (University of Bristol, GBR); Hetherington, Alexander (University of Bristol, GBR); Tarver, James (University of Bristol, GBR); Peterson, Kevin (Dartmouth College, Hanover, NH, USA)

Multicellularity is widely regarded as one of the most formative steps in evolutionary history and it has been made by many scions in the Tree of Life, however, the consequences have not been equal for all of the lineages that have made this transition. Most attention has been lavished on attempts to understand how animal bodyplans emerged from unicellular ancestors, apparently demonstrating not only that this was achieved very quickly, but that the extremes of disparity were achieved early. Causal hypotheses range from protein, gene regulatory,

and stochastic evolution. We attempted to test these hypotheses of pattern and process by establishing an empirical morphospace for the animal kingdom, the results of which demonstrate that while many clades exhibit maximal initial disparity, arthropods, chordates, annelids and mollusks have continued to explore and expand the limits of morphospace throughout the Phanerozoic, doubling the envelope of disparity occupied during the early phase of animal evolution. The morphological distances between phyla mirror differences in complexity, body size, and species-level diversity across the animal kingdom. A parallel analysis of the plant kingdom exhibits a pattern of increasing variance throughout evolutionary history. In testing hypotheses of causality, we find a strong correlation between increasing morphological disparity and genome size as well as disparities in microRNA repertoire, our proxy for gene regulation indicating potential mechanisms underpinning the emergence of multicellular organismal complexity in both the animal and plant kingdoms.

S14-03 Online databases provide critical insights into the evolution of appendage modularity during the fin to limb transition

Dececchi, Alex (University of South Dakota, Vermillion, SD, USA); Mabee, Paula (University of South Dakota, Vermillion, SD, USA); Marcot, Jonathan (University of Illinois, Urbana, IL, USA); **Sears, Karen** (University of Illinois, Urbana, IL, USA)

Morphometric and developmental studies suggest that the segments of the tetrapod limb (i.e., girdle, stylopod, zeugopod, and autopod) form distinct modules. This modular design is believed to allow the limb segments to evolve relatively independently, facilitating the adaptive evolution of the limb. Researchers have hypothesized that this modular pattern evolved during the fin to limb transition in response to the effect of changing functional pressures on the genetic architecture of the developing limb. However, this hypothesis has not been rigorously tested, and developmental data suggest that at least some of the genetic modules that regulate limb development in tetrapods were present in their finned ancestors. This raises the possibility that the modularity that characterizes the limbs of modern tetrapods may also have been present in more basal Sarcoptergians. To test among these hypotheses, a dataset of discrete limb characters was downloaded from Phenoscape. Phenoscape is a collaborative database designed to integrate developmental phenotypes with the fossil record. The downloaded database consisted of 39 published matrices from the Tetrapodomorph literature spanning the fin to limb transition. This dataset was then partitioned into four subsets, some of which overlap: all taxa, all tetrapods, all nontetrapods, and non-tetrapods that form the closest outgroups to tetrapods. In this study "tetrapod" refers to only those taxa with digits. For each dataset, the correlation of evolutionary changes between each pair of characters was calculated using the pairwise comparison method, Pagel's Test and Maddison's Test (implemented within a phylogenetic context in Mesquite and R), and the level of correlation within and among limb segments compared. Results indicate that character correlations are significantly higher within- than among- segments within all taxa and all tetrapods. In contrast, within- and among- segment correlations are similar in non-tetrapods. These results are consistent with an increase in the modularity of appendage segments around the time of the origin of the tetrapod limb, possibly in response to the limb's new functional role in terrestrial locomotion.

S14-04 Evolutionally conserved mechanisms of regeneration in chordates: Uncovering pathways active during WBR in *Botrylloides leachi*

Zondag, Lisa (University of Otago, Dunedin, NZL); Rutherford, Kim (University of Ontago, Dunedin, NZL); **Wilson, Megan** (University of Ontago, Dunedin, NZL)

Regenerative capacity differs greatly across organisms and the ability to regenerate declines as morphological complexity increases. Within the chordate phylum, vertebrate animals exhibits a very limited regenerative potential, however, the sea squirt *Botrylloides leachi* is a chordate with a remarkable ability to undergo whole body regeneration (WBR). A fully functional adult organism (zooid) can regenerate from a minuscule piece of vascular tissue within only 8 days. In order to compare the molecular mechanisms underlying WBR in a chordate to the regeneration process in other animals, we have analysed the transcriptome of B. leachi at each stage of regeneration. Following de novo transcriptome assembly (6 transcriptomes in total), differential expression analysis was performed to identify genes up or/and downregulated during WBR. Differential gene expression analysis indicates that both wound healing and an immune response are activated during early steps of regeneration in B. leachi WBR. These processes are also key to regeneration in response to injury in vertebrate models of regeneration such as the limb regeneation in salamanders. This suggests that chordate animals may employ a homologous series of molecular events during regeneration events wheather it be WBR or tissue regeneration.

11.10 – 12.50 Symposium S15:

Ecological and environmental impacts on the evolution of organismal development II

ROOM C1

Organizers: Chris Lowe, John Willis, and Angelika Stollewerk
Chair: Angela Stollewerk

S15-01 Evolutionary lability of plant epidermal morphology in response to changing interactions with animals

Glover, Beverley (University of Cambridge, GBR)

The enormous species diversity of the flowering plants has puzzled evolutionary biologists since Darwin's day. The rapid radiation of the flowering plants can be attributed at least in part to their recruitment of insects as vectors for their pollen, and the subsequent reproductive isolation that arose between populations with different floral morphologies. Understanding the relationships between flowers and their pollinators is important not only in understanding the rapid radiation of the angiosperms, but also in devising strategies to optimise yield of key insect-pollinated food crops such as fruits, legumes and oilseeds. In this talk I will discuss our recent research into how petal morphology influences pollinator behaviour and plant evolution. I will focus particularly on the petal epidermis, which is the primary point of contact between a flower and a pollinating animal. The morphology of petal epidermal cells influences flower function in a number of ways. Epidermal cell shape, position and surface patterning can alter flower temperature and scent, influence the way pollinators physically contact the flower, and produce a surprising array of optical effects including structural colours and iridescence. These various factors combine to determine how pollinating animals of different species behave when visiting the flower. We use a combination of approaches derived from the fields of molecular genetics and developmental biology, optics and material physics, systematics and evolutionary biology, and behavioural ecology and psychology to build an integrated picture of how the petal epidermis influences pollinator behaviour and ultimately plant reproductive success. I will describe how our work is beginning to define the molecular changes underpinning the evolutionary lability of epidermal morphology.

S15-02 New genes, new chemistry and new cells in phenotypic plasticity and the evolution of novelty in nematodes

Sommer, Ralf (Max-Planck Institute for Developmental Biology, Tübingen, GER)

My lab has developed the nematode *Pristionchus pacificus* as a model system for integrative studies in evolutionary biology by bridging eve-devo with ecology and population genetics. Studying phenotypic plasticity and its role for the evolution of novelty, we identified key roles for novel genes and novel small molecules at crucial positions in genetic networks. *P. pacificus* has a mouth dimorphism involved in alternative feeding strategies representing a case of phenotypic plasticity. Similarly, dauer development vs. direct development also represents a case of phenotypic plasticity. I will describe and report the results of our genetic and chemical studies that show how novel players are integrated into pre-existing genetic networks. Finally, I will describe a new regulatory function of a neuron for dauer formation that has not been observed in *C. elegans*.

S15-03 Understanding strategies used by the *C. elegans* reproductive system to cope with uncertain environments

Ruvinsky, Ilya (The University of Chicago, IL, USA)

To ensure long-term reproductive success in variable and largely unpredictable environments, organisms have to evolve ways of coping with uncertainty. For example, a simple reproductive strategy that maximizes offspring production under all conditions is likely to be disadvantageous because it will lead to a catastrophic loss of fecundity under unfavorable conditions. We seek to understand how an appropriate balance is achieved by investigated reproductive performance of *C. elegans* under conditions of chronic heat stress. I will discuss several recent results that may seem paradoxical. For instance, why do worms continue to reproduce under conditions in which none of their offspring can survive? Why greater stress can sometimes be less damaging than milder challenges? Why sex ratios can be both indicative and causal of different stress-coping strategies? These phenomena can be readily explained by considering the anatomy and physiology of the reproductive system and the trade-offs required to ensure reproduction under variable environmental conditions under which *C. elegans* evolved.

S15-04 Experimental evolution of multicellularity

Travisano, Michael (University of Minnesota, Saint Paul, MN, USA); Ratcliff, William (Georgia Institute of Technology, Atlanta, GA, USA)

The evolution of multicellularity dramatically changed life on earth, leading to vast changes in the complexity of life. Virtually all life that we can see without magnification is multicellular, and contains

from tens to trillions of cells. Investigating the evolutionary origins of multicellularity helps to understand the complexity of life, but is difficult, because multicellularity in nature evolved millions of years ago. To overcome this limitation, we've experimentally evolved multicellularity in the laboratory, starting with the single celled Baker's yeast. Using selection for fast settling in liquid, we show that multicellular yeast readily evolve from their single celled ancestor in as little as 7 days. The multicellular yeast grow by persistent attachment of daughter cells to mother cells, producing a characteristic — snowflake — body plan. These snowflake yeast evolve complex life-histories including juvenile and adult life stages, and respond to selection to body size. They also evolve a form of terminal cellular differentiation that promotes rapid growth. We've continued settling selection for over 7 months, and see that the multicellular yeast continue to increase in settling rate, and did so in three ways. First they increase in the number of cells as we initially observed, then they also increase the size of cells, and finally they evolve a more hydrodynamic shape. The first two multicellular adaptations make multicellular yeast bigger and thus faster settling. But larger size carries a cost: slower growth rates. The last adaptation, more hydrodynamic shape, allows for faster settling without imposing a growth rate cost. This suggests that the costs of increased size, which have been seen in algae and bacteria, may drive the evolution of increased complexity by favoring innovation.

11.10 – 12.50

Symposium S16:
How does developmental robustness facilitate the evolution of biodiversity?

ROOM C2

Organizers: Rainer Melzer and Günter Theissen
Chairs: Günter Theissen and Rainer Melzer

S16-01 How do clades explore morphological character space throughout their evolution?

Wills, Matthew (University of Bath, GBR); Hughes, Martin (Natural History Museum, London, GBR); Gerber, Sylvain (University of Cambridge, GBR); Oyston, Jack (University of Bath, GBR); Wagner, Peter (National Museum of Natural History, Washington DC, USA)

Extinct major clades diverge from a single common ancestor, subsequently describing some trajectory of diversity through time before their ultimate demise. Palaeontologists routinely study these trajectories, anecdotally observing that maximum diversity (numbers of species or genera) often occurs long after a clade's inception. Surprisingly, however, the early history of a group is often characterized by species of greater morphological distinctiveness – greater morphological disparity – than subsequently, despite relatively low diversity at this time. A sample of 63 clades not going extinct coincident with one of the "big five" mass events ("freely evolving" clades) demonstrated a significant bias towards early high disparity; a low centre of gravity in disparity profile. We briefly consider the extent to which character exhaustion and subsequent convergence offers an explanation for the apparent limits on the further morphological expansion of such groups. By contrast, a sample of 35 clades terminating at a mass extinction event showed a significant bias towards late high disparity, similar to a sample of 53 extant clades with disparity profiles truncated by the Recent. Clades radiating in the immediate wake of a mass extinction do not appear to show early high disparity, suggesting that morphological diversification may be impeded by ecosystem collapse (rather than induced by empty ecospace).

S16-02 The evolution of canalization and evolvability in changing environments

Hansen, Thomas F. (University of Oslo, NOR)

Waddington hypothesized that canalization (robustness) would evolve under stabilizing selection in a stable environment. It is often thought that changing or fluctuating environments would have the opposite effect, favoring decanalization and increased evolvability, and thus allowing more rapid adaptation in changing environments. In this presentation I analyze this situation with theoretical models. I show that the situation is not so simple. Directional selection can act canalizing or decanalizing depending on patterns of epistasis, and fluctuating selection induces some the same canalizing selection pressures as stabilizing selection. More generally, I argue that the evolution of variational properties such as canalization, robustness, modularity and evolvability is mainly governed by indirect selection due to correlations with trait means, and is as dependent on the shape of the genotype-phenotype map as on the pattern of selection.

S16-03 Developmental robustness in *C. elegans*: From quantification to mechanisms

Barkoulas, Michalis (Imperial College, London, GBR); Felix, Marie-Anne (Ecole Normale Superieure, Paris, FRA)

Biological systems are continuously subject to intrinsic or extrinsic variation, such as genetic variation due to mutation, stochastic variation in the abundance of critical macromolecules and environmental variation in growth conditions, yet they often manage to operate with remarkable precision akin to the most optimally engineered machines. The property of a biological system to produce

an invariant output in the presence of considerable variation is called robustness. Development itself is highly robust to noise and this is instrumental for the successful transformation of a fertilised egg into a multi-cellular individual. Developmental robustness ensures the stability of phenotypic traits, including correct cell numbers in a given tissue. While research on developmental robustness has seen a surge over the last 15 years, most studies have remained theoretical, and we still lack appropriate experimental systems to elucidate the mechanisms via which robust outputs are achieved. We use the stereotypical patterning of isogenic *C. elegans* as a model to study the mechanisms of developmental robustness and how these mechanisms evolve. I will present our efforts to quantify at single molecule resolution the boundaries of developmental robustness to pathway dose variation in the *C. elegans* vulva. I will show that gene expression of the master-inducer of the vulva is robust to vast genetic diversity in *Caenorhabditis* and turnover of *cis*-regulatory sequences. I will finally present how by focusing on a new experimental model, the epithelial seam cell number variation, we hope to derive principles about the contribution of genes to stabilisation of developmental outcomes and develop a developmental framework for phenotypic buffering.

S16-04 Genetic basis of petal number variation in *Cardamine hirsutas*

Hay, Angela (Max Planck Institute for Plant Breeding Research, Cologne, GER)

A key question in biology is how differences in gene action produce morphological variation during evolution. *Cardamine hirsuta* — a plant related to the model organism *Arabidopsis thaliana* — is a powerful experimental system for investigating this question. *C. hirsuta* has diverged from *A. thaliana* in a number of key morphological traits, including petal number. Petal number is constant in *A. thaliana* but variable in *C. hirsuta*. Using genetics and reciprocal cross-species gene transfers we found that *cis* regulatory divergence in a single gene is sufficient to account for petal number differences between the two species. We found that distinct quantitative trait loci (QTL) explained natural variation in *C. hirsuta* petal number, demonstrating thatinter and intra-specific variation in petal number has a different genetic basis. Intriguingly, we found that these QTL effects are conditional on the gene underlying species-specific petal number. Thus, our results show how a single gene contributed to the evolution of petal number variation in *C. hirsuta* by shifting this trait from an ancestral robust state into a state in which petal number is no longer buffered, and has thus become subject to genetic and environmental perturbation.

14.20 – 16.00 Symposium S17: EvoDevo as an approach to understanding communication

ROOM A

Organizers: D. Kimbrough Oller and Ulrike Griebel
Chair: D. Kimbrough Oller

S17-01 Language evolution and change: The impact of modern evolutionary thinking

Dediu, Dan (Max Planck Institute for Psycholinguistics, Nijmegen, NLD)

The origins and evolution of language and speech, and the processes governing language change represent major areas of interdisciplinary research impacting not only on language sciences but also on the fundamental question of what it means to be human. However, their scientific investigation is notoriously difficult due to a lack of data and, partly as a consequence, a tendency for debates to be driven by a priori strong theoretical positions. Probably the dominant proposal within linguistics was that language emerged suddenly and recently, coincident with a speciation event that resulted in our own species, *Homo sapiens*. Various proposals included a single (or a few) genetic mutation(s) resulting in the sudden appearance of core language properties (such as recursion) through unspecified mechanisms, sometimes with clear anti-evolutionary connotations. However, recent advances in our understanding of language and speech and their neural and genetic underpinnings strongly advocate against such saltationist scenarios, and together with new data from archaeology, palaeoanthropolgy and ancient DNA, favour the gradual emergence of language and speech on a much longer timescale going back at least to our last common ancestor with the Neandertals. The complexity of these biological foundations of language require new theories of language acquisition and use that highlight the constant interaction between culture and genetics. Another important insight is represented by the role played by language as a cultural phenomenon in a process of gene-culture co-evolution whereby language constructs a specific niche which, in turn, changes the landscape of selective pressures acting on our genome. In this sense, language and speech (and the culture they support) are very powerful cases of phenotypic plasticity with trans-generational consequences. Relatedly, the idea that language is a true evolutionary system in itself becomes more and more mainstream, and methods adapted from evolutionary biology (such as Bayesian phylogenetics and phylo-geography) are successfully applied to recalcitrant problems in historical linguistics. Thus, recent advances such as evo-devo, the appreciation of phenotypic plasticity and developmental robustness and the complexity of the evolutionary processes afforded by the structure of our genomes, both inform and can be informed by debates in language evolution. However,

we must be careful in how we transfer such concepts, methods and findings across disciplines, as there is the real danger that superficial and metaphorical appeals to evo-devo are used to justify old and fundamentally non-evolutionary proposals, confusing the literature and creating false debates.

S17-02 The building blocks of language: From molecules to neuronal circuits

Vernes, Sonja (Max Planck Institute for Psycholinguistics, Nijmegen, NLD)

The capacity for language is a fundamental trait unique to humankind. As such, studying the neurogenetic substrates of language presents a major scientific challenge. The capacity for language is likely to have arisen from adaptation of existing genes and brain structures. Thus, animals can be used to model evolutionarily conserved pathways and understand neurogenetic mechanisms that may have contributed to the development of language related circuits in the brain. An excellent example of the success of this strategy is given by the *FOXP2* gene. *FOXP2* was the first gene implicated in human language due to its identification in a large Mendelian pedigree of speech and language impairment (the KE family). Study of *FOXP2* in cellular and animal models has provided insight into how disruption of a single gene that is expressed in a range of tissues during development, can lead to a specific disorder affecting language related phenotypes. This work has also opened the door to wider investigations of whole molecular pathways, leading to identification of new candidate genes in other language related disorders and molecular links between clinically distinct syndromes. Furthermore, advances in electrophysiological and imaging technologies have allowed investigation of the roles of these molecular networks at the level of the neuronal network and the whole brain. I will discuss recent developments in language genetics from the level of the single gene, to individual cells, and neuronal networks in an effort to bridge the gap between genes, brains and communicative behaviour. By using a multi-level approach such as this, we can begin to understand the molecular and cellular building blocks underlying the evolution and development of complex behaviours like language.

S17-03 Computational models of human vocal development and evolution

Warlaumont, Anne (University of California Merced, CA, USA)

Because of the complexity of the neural, physical, and social underpinnings of human communication, the ethical constraints on experimenting directly with human children, and the sparsity of direct evolutionary evidence, computational modeling provides a particularly useful approach for studying the development and evolution of human vocal signals. This presentation will review recent accomplishments in simulating the earliest stages of emergence of human vocal communication and then suggest some particularly important future directions. A number of computational models have been built to account for how young children learn to produce speech-like sounds. These models have demonstrated that both endogenous and social factors can contribute to vocal learning. Endogenous factors include an interest in auditorily salient sounds and a natural curiosity and desire to learn about the perceptual effects of one's actions. Social factors include responding by adults that is contingent on the types of sounds the chid produces, and providing examples of adult speech as input to the child. These models have been developed at a range of physiological detail, with some including physical models of the human vocal tract and some using simulated neural networks to perform the learning. Modeling at this level of detail sheds light on how vocal learning may be supported and constrained by the physics of of the sound-generating mechanism and by the computational properties of neural networks. Additionally, some of this modeling work has shown how differences intrinsic to children, such as differences in vocalization rate, and differences in the individuals the child interacts with, such as differences in adult response rates, lead to differences in child speech development that match those seen in human children growing up in different socioeconomic environments and in children with or without autism. The work thus points to possible evolutionary changes that affected the development of human vocalization, in turn affecting the nature of vocal communication in adult humans. Nevertheless, more work directly targeted at the evolution of human vocal development is needed. In particular, future models should combine genetic adaptation and developmental adaptation (via learning) in both children and caregivers, informed by both genetic and behavioral human data. In addition, models focusing on the relationship between human speech-related vocalizations and phylogenetically older human vocal signals such as screams, laughs, and cries would help address the question of how human speech evolved.

S17-04 Human vocal development and animal communication in an EvoDevo approach to language

Oller, D. Kimbrough (University of Memphis, TN, USA); Griebel, Ulrike (University of Memphis, TN, USA)

Human and animal communication show huge differences in complexity and flexibility. By the first three months of life human infants produce non-cry "protophones" (squeals, growls, raspberries…) which can be used with full functional and contextual flexibility in systematic face-to-face vocal interaction with caregivers, who imitate and adjust their infant-directed speech to include acoustic

features of the protophones. The endogenous vocal exploratory tendency and the social interaction tendency create an interplay that cyclically builds foundations for language. Interaction with caregivers provides a cultural support system and affords selection pressures on these foundations. As the first year proceeds the interactions become more elaborate, laying additional foundations. Such patterns of infant vocalization and sustained face-to-face vocal interaction with caregivers have never been reported in non-human primates. Animals in the wild have far less elaborate communication systems than humans, with no evidence of true lexicon or true syntax. Yet many animals can learn aspects of language with intense human enculturation. Animals, including great apes, have been shown in several instances to learn to recognize hundreds of lexical items and to retrieve hundreds of objects named by humans. In addition, great apes have learned to produce lexical items that possess at least limited semanticity and to combine such lexical items in primitive propositions/syntax. These language-trained apes have also learned to communicate lexically about events and objects that are not in the here-and-now. Yet the extent of animal communicative capabilities in the wild is vastly different from the capabilities seen after human training. The phenotypic variation within these human-trained animals shows that communication capabilities are very sensitive to "cultural" environment, making it possible for some animals to adapt communicatively far beyond the typical boundaries seen in their species. The results of these studies indicate shared cognitive capabilities relevant for language learning across many species. Nevertheless, the human infant's plasticity with respect to language is far greater. By 36 months, typical human infants have been enculturated linguistically far beyond levels seen in any animal training study — spoken vocabularies often reach thousands and receptive vocabularies many thousands, with sentences composed of up to 20 words, and children are able to talk about events in the present, the past, the future, and the imagination, often in more than one language. A modern evodevo approach to language should address the steps, beginning in the first month of human life that lead through enculturation to vast phenotypic plasticity.

14.20 – 16.00 **Contributed Session C11:**
What should Bioinformatics do for EvoDevo?

ROOM B

Chairs: Günter Plickert and Paula Mabee

C11-01 Phylogenomics of MADS-box genes in flowering plants to identify EvoDevo genes

Theissen, Guenter (Friedrich Schiller University Jena, GER); Gramzow, Lydia (Friedrich Schiller University Jena, GER)

Identifying developmental control genes and assessing their evolutionary importance is one aspect of what Bioinformatics can do for EvoDevo. We use MADS-box genes in flowering plants to illustrate this point. MADS-box genes control many processes in flowering plants, such as flower and fruit development. In the genomes of flowering plants, on average about 100 different MADS-box genes can be found. These genes were classified into five major groups that were further subdivided into ancient clades ("subfamilies"). Here, we study the 17 clades of MIKCc-group MADS-box genes that had already been established in the most recent common ancestor (MRCA) of extant flowering plants more than 150 million years ago. For each of these clades we determine the presence or absence in the whole genomes of 27 flowering plant species. We find that all members of two clades (TM8- and FLC-like genes) have been lost in multiple species, suggesting that the function of these genes is dispensable in some lineages or that gene loss is even adaptive under some circumstances. In contrast, a number of clades have never been wiped out completely in any of the investigated species. Surprisingly, these highly conserved clades do not only include the ones for which developmental functions have been well defined based on dramatic mutant phenotypes of some clade members, but also four other clades (TM3-, StMADS11-, AGL17- and GGM13-like genes) whose developmental importance is by far less clear. Our data suggest that the genes in these clades are generally more important for angiosperm development and evolution than has previously been appreciated.

C11-02 Illuminating the evolutionary origin of the turtle shell by a comparative tissue-specific transcriptome analysis

Pascual-Anaya, Juan (RIKEN Center for Developmental Biology, Kobe, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

The turtle shell is a genuine morphological innovation within tetrapods. Its formation requires a complete anatomical distortion of the tetrapod body plan, resulting in an open, fan-like ribcage formed by plate-like ribs that eventually enclose the shoulder girdle (otherwise, remaining outside in the rest of amniotes). During development, the turtle shell is preceded by the formation of an ectodermal ridge, underlain

by a condensed mesenchyme, running anterior-posteriorly through the dorsal flank of the body in the inter-limb region, the so-called carapacial ridge (CR). The CR is thought to control the development of the shell. In recent studies, we have shown the specific expression of key developmental genes in the CR, not expressed in the dorsal flank of chicken neither in the body wall of the turtle. These genes might have important functions in the shell development, at least regarding the correct fan-like disposition of the ribs. These genes have probably been co-opted from other developmental modules (such as the limbs, as first proposed by Anne Burke in 1989). However, the whole gene regulatory network(s) controlling the formation of the CR remains a mystery. In this study, we have analyzed the transcriptomes of the CR and other structures (limbs and body walls) from the Chinese soft-shell turtle, *Pelodiscus sinensis*, and compare it with the transcriptomes of homologous or equivalent embryonic regions of the chicken (dorsal flank, limbs and body walls). Our (inter and intra-species) comparative analyses allowed us to identify those genes that are expressed in the CR of *P. sinensis*, and that might be involved in the turtle shell development. We next coupled our transcriptomic data with a preliminary ChIP-seq analysis for histone modifications of the CR and other tissues, in order to depict the putative genomic changes at the regulatory level accompanying the co-option of these genes to the CR from previously unrelated developmental modules.

C11-03 Blastodermal segmentation in the milkweed bug, *Oncopeltus facsiatus*

Chipman, Ariel (The Hebrew University of Jerusalem, ISR); Stahi, Reut (The Hebrew University, Jerusalem, ISR)

The insect segmented body plan is conserved and stereotypical. However, it is becoming increasingly clear that the embryonic processes leading to this conserved body plan are highly variable. Long germ segmentation, best known from *Drosophila melanogaster*, is characterized by a complex hierarchy of gene interactions that results in the generation of all segments during the blastoderm stage. This is the more evolutionary derived mechanism, and it can be found in most holometabolous insects. Short germ segmentation (or sequential segmentation) is characterized by a regulatory network producing clock-like oscillations that produce most trunk segments after gastrulation. Sequential segmentation is the ancestral mechanism, and is the predominant process in all insects outside of the Holometabola. However, the segmentation process in most insects actually uses a mix of both mechanisms. The anterior-most segments (normally the head and thorax) appear more or less simultaneously during the blastoderm stage, while the rest of the segments (normaly abdominal segments) appear sequentially from a growth zone, during the germ-band stage.

In order to understand how long germ development evolved from the ancestral short germ pattern, we have been looking at the hemipteran *Oncopeltus fasciatus*. The Hemiptera are the closest hemimetabolous outgroup to Holometabola, making them a suitable comparison to the well-studied holometabolous development pattern. Our model species, *O. fasciatus*, displays intermediate germ segmentation, which includes a blastoderm stage similar to that of *D. melanogaster*, and a growth zone with sequential segmentation. We ask whether blastoderm segmentation in *O. fasciatus* is similar to the sequential segmentation from its growth zone or to the simultaneous segmentation seen in *D. melanogaster*. We have characterized the expression, dynamics and function of four segmentation genes: *engrailed*, *wingless*, *even-skipped* and *delta*, during blastoderm stages of *O. fasciatus*. In addition, we have looked at the role of gap gene orthologs during blastodermal segmentation to try and understand if they are necessary for anterior segment generation or only for determining their identity.

C11-04 The origins of arthropod innovations: Insights from the non-insect arthropods, the cherry shrimp and rusty millipede

Kenny, Nathan (The Chinese University of Hong Kong, HKG); Qu, Zhe (The Chinese University of Hong Kong, HKG); Wong, Nicola (The Chinese University of Hong Kong, HKG); Lam, Hon Ming (The Chinese University of Hong Kong, HKG); Chu, Ka Hou (The Chinese University of Hong Kong, HKG); Hui, Jerome (The Chinese University of Hong Kong, HKG)

For the past century insects have played a leading and vital role in developmental, and, more recently, EvoDevo research, due to their rapid generation times and ease of culture in the laboratory. This dominance has only increased with the advent of genomic sequence data, and has risked skewing our perception of the drivers of phenotypic change. As insects are but a single (but admittedly numerous) clade in the diverse Metazoa, whether the patterns and processes studied in the Insecta are truly representative of animal life as a whole is proving increasingly contentious. The advent of NGS technology has meant these hypotheses can now be tested in non-Insect, non-model species. Furthermore, we have had a tendency to catalogue novelties on the basis of our limited sampling of Metazoan diversity. The rise of non-model genomic resources will mean we can truly determine the origin of novelties, and more precisely track the gain and loss of adaptive phenotypes across animal phylogeny. Sequencing the genomes of two non-insect arthropods, the cherry shrimp *Neocaridina denticulata* and rusty millipede, *Trigoniulus coralinus*, we are testing the origins and evolution of a number of developmental gene and microRNA families. By leveraging the advantages of NGS technologies in this manner the true reasons for the success of the diverse and speciose Arthropoda can be gained.

14.20 – 16.00 Contributed Session C12: Perspectives on Wnt signaling II

ROOM C1

Chair: Wim Damen

C12-01 Repeated evolution of novel embryonic axis determinants in dipteran insects

Schmidt-Ott, Urs (The University of Chicago, USA)

Establishing the primary body axis is fundamental to animal development. Anteriorly localized mRNA transcripts of the homeobox gene bicoid guide the formation of this axis in embryos of the fruit fly *Drosophila melanogaster*. However, in other insects the mechanisms underlying head-to-tail polarity are not understood. We identified a novel gene, panish, that establishes head-to-tail polarity in a close relative of biting mosquitoes, *Chironomus riparius*. *Panish* encodes a small protein with a cysteine-clamp DNA binding domain similar to that of the Wnt signaling effector Pangolin but is otherwise completely different and may best be viewed as a novel gene because it also shares sequence with another protein. Maternal *panish* transcripts are enriched in the anterior syncytial embryo and knockdown induces a double-abdomen phenotype that can be rescued by injection of *panish* mRNA. The *panish* gene is absent in other fly species, indicating repeated evolution of long-range embryonic pattern organizers in dipteran insects. Ongoing research on the functional evolution of *panish* will be reported.

C12-02 Antagonizing Wnt signaling in the Tribolium embryo

Schröder, Reinhard (University of Rostock, GER); Prühs, Romy (University of Rostock, GER); Sharma, Rahul (University of Rostock, GER); Beermann, Katharina (University of Rostock, GER); Beermann, Anke (Eberhard Karls University of Tübingen, GER)

During embryogenesis of animals, Wnt-signalling is essentially required for various processes such as body axis elongation, limb development, organogenesis and stem cell proliferation. Yet, Wnt signalling has to be carefully controlled by modulators and inhibitors to prevent excess and ectopic Wnt activity that would negatively interfere with normal development and can lead to cranial defects or to cancer. Here, we analyse the role of two Wnt-inhibitors Tc-LRP4 and Tc-Axin during embryogenesis of the beetle *Tribolium*. LRP4 is thought to inhibit Wnt signalling in a dominant-negative way by competing with Arrow/Lrp5 for Wnt ligand binding. In Tribolium, Tc-lrp4 is ubiquitously expressed. When interfering with Tc-lrp4 function embryos develop to larvae with reduced head structures, constrictions and large gaps. Tc-axin as a member of the beta-Catenin destruction complex is involved in keeping the level of cytoplasmic beta-Catenin low. In contrast to Tc-lrp4, Tc-axin is distinctly expressed in various tissues throughout *Tribolium* development. When knocked down by RNAi, Wnt-activity is flattened at a high level within the egg. As a consequence, posterior structures expand at the expense of head- and thoracic development. Marker gene expression studies of Tc-lrp-RNAi and in Tc-axin-RNAi embryos revealed the expanded expression of the Wnt-target Tc-caudal and of segmentation genes illustrating the consequences of unwarranted high levels of Wnt-activity during embryogenesis.

C12-03 Wnt-Myc interaction in Hydra stem cell proliferation

Hobmayer, Bert (University of Innsbruck, AUT); Gufler, Sabine (University of Innsbruck, AUT); Glasauer, Stella (University of Innsbruck, AUT); Bister, Klaus (University of Innsbruck, AUT); Hartl, Markus (University of Innsbruck, AUT)

Wnt signaling has been demonstrated to act in axial patterning and regeneration throughout the metazoans. In higher animals, it also has been shown to regulate cell proliferation in embryonic tissues and tumor cells. Myc, on the other hand, represents one of the best-studied cell cycle regulators due to its progressive role in human tumors. Recently, we have started to characterize two members of the *myc* proto-oncogene family in the cnidarian polyp *Hydra* in order to trace ancestral Myc functions (Hartl et al. 2010, 2014). Both genes encode for prototypic Myc proteins showing substantial structural conservation, Max dimerization, E-box-specific DNA binding and a basic competence for oncogenic transformation when tested in avian fibroblasts. Both genes are activated in proliferating cells of the interstitial stem cell system of *Hydra*. siRNA mediated knockdown of myc1 results in enhanced interstitial stem cell proliferation (Ambrosone et al. 2012). Here, we demonstrate that both genes are also transcriptionally regulated by Wnt/beta-Catenin signaling. Activation of beta-Catenin using pharmacological agents or using stable beta-Catenin transgenic polyps results in down-regulation of myc1 and in moderate up-regulation of *myc2*. ChIP experiments show binding of Tcf at specific sites in the *myc1* and *myc2* enhancers. Assays using *Hydra* beta-Catenin and *Hydra* Tcf overexpression constructs in combination with a Luciferase reporter confirm down-regulation of *myc1* and up-regulation of *myc2* in vertebrate cell culture. In summary, our data provide a direct link between position-specific Wnt/beta-Catenin signaling and Myc factors in stem cell proliferation control in a simple animal model.

C12-04 How regulatory beta-catenin modules impinge on early annelid GRNs

Schneider, Stephan (Iowa State University, Ames, IA, USA); Pruitt, Margaret (Iowa State University, Ames, IA, USA); Bastin, Benjamin (Iowa State University, Ames, IA, USA); Chou, Hsien-chao (Iowa State University, Ames, IA, USA)

Although conserved features among developmental gene regulatory networks (GRNs) between various deuterostome embryos are emerging, a comparison to protostomes remains difficult due to the highly derived nature of early embryogenesis and the genomes in the protostome models *C. elegans* and *Drosophila* (Ecdysozoa). To fill this gap we focus on early developmental mechanisms in the protostome *Platynereis dumerilii* (Annelida, Lophotrochozoa). *Platynereis* has retained a more ancestral gene set without the extensive gene loss and gain observed in the other protostome models. In addition, early development of *Platynereis* is attractive as it exhibits a series of invariant, stereotypic asymmetric cell divisions, allowing individual embryonic cells to be recognized by size and position. Furthermore, *Platynereis* exhibits a global reiterative beta-catenin mediated cell fate specification mechanism in early development that appears to convey lineage-specific binary cell fate decisions. To gain insights into the early embryonic gene regulatory networks (GRNs), and the contribution of reiterative beta-catenin asymmetries to specify cell fates, we deployed a variety of RNA-seq based approaches. Comparing normal embryos, and embryos with ectopically induced beta-catenin mediated cell fate transformations, we have identified transcriptional changes during early development from the zygote through the mid gastrula (1 cell stage through ~330 cell stage), and after distinct cell fate transformations. We have subsequently mapped the expression of developmental regulators including beta-catenin pathway components (ligands, receptors, transcription factors) into distinct cell lineages. Our approaches capture stage-specific transcriptional snapshots of normal and manipulated *Platynereis* embryos and thereby provide the first view of early embryonic GRNs that utilize a spiral-mode of cell divisions to segregate cell fates, and how beta-catenin regulatory modules impinge on early annelid GRNs.

14.20 – 16.00 Contributed Session C13: Ecological and environmental impacts on the evolution of organismal development I

ROOM C2

Chair: Angelika Stollewerk

C13-01 Combining molecular, developmental, and ecological approaches to understanding the relationship between genotype, phenotype, and the selective environment

Khila, Abderrahman (Institute of Functional Genomics of Lyon, FRA)

Understanding the ecological, evolutionary, and molecular mechanisms underlying morphological diversification is a major challenge in biology. A combination of historical, biological, and technical features establishes the semi-aquatic insects as a sustainable model for integrating various approaches to the study of animal diversity. In water striders, the length and shape of locomotory appendages are critical not only for water surface locomotion, but also for escape strategy from bottom striking predators such as fish. In this talk, I will first detail a concrete example of how gene interaction can shape an adaptive morphological trait during development and evolution. Then I will describe why this specific phenotype has been favored by natural selection, within the context of prey-predator interaction. Finally I will show how genetic manipulation of this phenotype can affect escape performance of the prey, and therefore reduce individual fitness. Integrating evo-devo with evolutionary ecology allows for a comprehensive and thorough understanding of phenotypic evolution.

C13-02 Evolution of the olfactory sensory system in the blind cavefish *Astyanax mexicanus*

Retaux, Sylvie (CNRS, Gif-sur-Yvette, FRA); Bibliowicz, Yoni (CNRS, Gif-sur-Yvette, FRA); Hinaux, Hélène (CNRS, Gif-sur-Yvette, FRA); Blin, Maryline (CNRS, Gif-sur-Yvette, FRA); Alié, Alexandre (CNRS, Gif-sur-Yvette, FRA); Espinasa, Luis (Marist College, Poughkeepsie, NY, USA)

We use the emerging fish model *Astyanax mexicanus* to understand genetic and cellular mechanisms involved in morphological and behavioral evolution. Within this species, there are populations of river-dwelling surface fish ("normal fish") and populations of blind cavefish inhabiting the darkness of caves. The latter have undergone some striking losses (eyes, pigmentation) but have also undergone some probably adaptive gains, such as more taste buds, more neuromasts, larger jaws, and more teeth. Their physiology and behavior is also very different from their surface counterparts. Here we have performed a comparative analysis of olfactory system development in cavefish and surface fish: early placodal patterning (*Dlx3b, Eya2,*

OMP), and proliferation patterns (PCNA, PH3) and neurogenesis (EdU incorporation) patterns in the olfactory epithelium appear different in cavefish, and result in a significant difference in olfactory organ size between the two morphs. In parallel, we have assessed the olfactory capabilities of the two *Astyanax* morphs, both in the wild and in the lab. Experiments performed in the Subterráneo cave (Mexico) show that cave morphs strongly respond to food-related olfactory cues. Experiments performed in the lab in controlled conditions and using amino-acids as olfactory cues show that cavefish have better olfactory discrimination threshold than surface fish. We propose that developmental evolution of the olfactory sensory system confers better olfactory capabilities to cavefish. Such a change can probably be considered as adaptive in response to the extreme dark environment where cavefish live.

Our work is supported by ANR (ASTYCO and BLINDTEST) and FRC144.

C13-03 Optogenetics illuminates the neural circuit regulating swimming behavior in the marine annelid *Platynereis dumerilii*

Tosches, Maria Antonietta (European Molecular Biology Laboratory, Heidelberg, GER); Bucher, Daniel (European Molecular Biology Laboratory, Heidelberg, GER); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER)

Animal behaviour evolves and adapts to the environment. Behaviour is the output of the activity of networks of neurons, which process sensory information and coordinate motor output. Therefore, to interpret nervous system diversity and evolution at different scales it is essential to integrate development and morphology with an understanding of how neural circuits function and evolve across phylogeny. Planktonic ciliated larvae are of special interest for studying the origin of nervous systems. Larval neurons respond to a variety of sensory stimuli and control swimming by innervating locomotor ciliated cells. Despite the apparent simplicity of these larval nervous systems, the neural circuits underlying sensory-motor integration in ciliated larvae are poorly understood. Using the annelid *Platynereis dumerilii* as a model, we dissected the mechanism that controls ciliary arrests and that modulates larval swimming under the influence of ambient light. To this aim, we first established in *Platynereis* calcium imaging of neuronal activity, using the optogenetic calcium indicator GCaMP6 and two-photon microscopy. This allowed the identification of a pair of cholinergic ciliomotor neurons in the larval brain, which innervate the locomotor ciliated cells and trigger ciliary arrest. Interestingly, electrophysiology and calcium imaging revealed that during the night the ciliomotor neuron changes its firing properties: it switches from single, sparse firing events to a rhythmic activity, characterised by the regular discharge of bursts of action potentials. This change in firing mode has a facilitatory effect on synaptic transmission; consistently, the rate of ciliary arrests is enhanced during the night. Next, we found that melatonin, released during the night by non-visual photoreceptors, controls directly the activity of the cholinergic ciliomotor neurons. Melatonin receptors are expressed in the ciliomotor cholinergic cells, and melatonin application during the day is sufficient to induce rhythmic firing and to recapitulate the nocturnal behaviour. Given the widespread occurrence of cholinergic ciliomotor neurons in marine animals, a similar circuit organisation could explain how in simple larval brains sensory inputs are integrated to control swimming. Moreover, we propose that the melatonin-dependent modulation of neuronal activity contributes to the rhythmic vertical swimming of zooplankton in the ocean, known as diel vertical migration.

C13-04 Genetic an epigenetic bases of abdominal pigmentation plasticity in *Drosophila melanogaster*

Gibert, Jean-Michel (UMR7622 CNRS-UPMC IBPS, Paris, FRA); Mouchel-Vielh, Emmanuèle (UMR7622 CNRS-UPMC IBPS, Paris, FRA); De Castro, Sandra (UMR7622 CNRS-UPMC IBPS, Paris, FRA); Coulpier, Fanny (Genomic Paris Centre, IBENS, Paris, FRA); Le Crom, Stéphane (Genomic Paris Centre, IBENS, Paris, FRA); Peronnet, Frédérique (UMR7622 CNRS-UPMC IBPS, Paris, FRA)

Environmental conditions can strongly modulate the phenotype produced by a given genotype. This phenomenon, called phenotypic plasticity, plays a major role in the wild where environmental conditions vary spatially and temporally. Furthermore, it is thought to facilitate evolution by revealing cryptic genetic variation and by genetic assimilation. However, its genetic bases are not well understood. We use as model of phenotypic plasticity the pigmentation of the posterior abdomen in *Drosophila melanogaster* females. This trait is strongly modulated by temperature. Our goal is to understand how temperature affects the gene regulatory network controlling posterior abdominal pigmentation, in particular via an effect on the transcriptome and the epigenome. We have compared the transcriptome of the abdominal epidermis from young isogenic females raised either at 18°C or 29°C (RNAseq). We found that, among other genes, *tan*, encoding an enzyme involved in melanin production, is 12 times more expressed at 18°C than at 29°C. Moreover, the two genes neighboring *tan* are also modulated by temperature, suggesting that *tan* belongs to a chromatin domain sensitive to temperature. Modulation of *tan* expression by temperature seems essential for pigmentation plasticity, as down-regulating *tan* at 18°C mimics the effect of heat. Using a GFP transgenic reporter line, we have shown that the effect of temperature on tan expression is mediated at least partly by an enhancer driving *tan* transcription in posterior abdominal epidermis. We are currently studying histone marks on *tan* promoter

and enhancer, in order to see whether some of them are modulated by temperature. Furthermore, we are performing RNAseq experiments on pupae to identify genes involved more precociously in phenotypic plasticity of female abdominal pigmentation.

14.20 – 16.00 Contributed Session C14: Developmental mechanisms underlying evolutionary change II

ROOM D

Chair: Peter Dearden

C14-01 A mechanism for reproductive constraint in the honeybee

Duncan, Elizabeth (University of Otago, Dunedin, NZL); **Dearden, Peter** (University of Otago, Dunedin, NZL)

Eusociality depends on reproductive division of labour. This implies that one individual, the queen in honeybees, can repress reproduction in other females. In Honeybees this is achieved through queen bees producing a pheromone, queen mandibular pheromone, which causes worker bees to repress the activity of their ovaries, repressing worker reproduction. Removal of the queen, and thus queen mandibular pheromone, allows the workers to activate their ovaries and begin to lay eggs. We have examined changes in gene expression, using high-throughput sequencing, in worker ovaries as they respond to the absence of queen mandibular pheromone and become activated. Using this dataset we have investigated the mechanisms by which queen mandibular pheromone has its action. While it is not clear how queen mandibular pheromone is detected, or how its activity is transduced, we have identified a cell-signalling pathway that responds to the absence of queen pheromone, and, when blocked, enhances the activation of the ovaries. Comparison of the action of this pathway in the presence or absence of queen pheromone implies that this is the main pathway by which the activity of worker ovaries is constrained in the honeybee, giving us insights into both the mechanism and evolution of reproductive constraint.

C14-02 EvoDevo of *Astyanax mexicanus* cavefish: A new time frame and its consequence on the underlying evolutionary mechanisms

Fumey, Julien (LEGS, UPR9034 CNRS, Gif-sur-Yvette, FRA); Noirot, Céline (Genotoul, INRA, Auzeville, FRA); Hinaux, Hélène (Neurobiology & Development Laboratory UPR3294, Gif-sur-Yvette, FRA); Rétaux, Sylvie (Neurobiology & Development Laboratory UPR3294, Gif-sur-Yvette, FRA); **Casane, Didier** (LEGS, UPR9034 CNRS, Gif-sur-Yvette, FRA)

Populations of blind cavefish belonging to the Mexican tetra species *Astyanax mexicanus* are outstanding models to study the evolution of vertebrates at a small time scale. In particular, the phenotypic convergence of independently evolved, cave-adapted populations allows questioning whether the evolution of similar cave phenotypes involved the fixation of standing genetic variation or the apparition of de novo mutations. To get an estimation of the time frame of the evolution of the *Astyanax* Pachón cave population, which is considered as one of the "oldest" and most isolated populations in the Sierra del Abra, we applied a population genomics approach. We compared transcriptome-wide the polymorphism and substitution rates of the Pachón population and a surface fish population (San Solomon Spring, Texas), using the Buenos Aires tetra (*Hyphessobrycon anisitsi*) as a close outgroup to identify ancestral and derived alleles. These data were compared to simulations of population evolution in which various parameters varied, such as the size and age of the populations and the gene flow between populations, to determine parameters that are compatible with the differentiation observed. The polymorphism was higher in the surface population than in the cave population, suggesting, as expected, a higher effective population size for the river-dwelling fish population. We also observed higher substitution rates in cavefish than in surface fish, also in accordance with a lower cavefish population size allowing a more rapid fixation of derived alleles, but implying that the Pachón cave population is much "younger" than previously estimated and may have spent less than 100.000 years underground. We will discuss the consequences of this new time frame on the underlying evolutionary mechanisms responsible for the morphological changes observed in cavefish populations.

Work supported by ANR [BLINDTEST].

C14-03 Notch signalling is necessary for environmentally-induced transdifferentiation in the sponge, *Amphimedon queenslandica*

Degnan, Bernie (University of Queensland, Brisbane, AUS); Nakanishi, Nagayasu (University of Queensland, Brisbane, AUS); Richards, Gemma (University of Queensland, Brisbane, AUS)

Metazoans rely on Notch signalling to an extraordinary extent. It functions in a wide range of developmental and physiological contexts, and thus influences many cellular decisions over the life of an animal. Notch also is involved in conferring plasticity onto terminally differentiated cells and their conversion in regeneration and tumorigenesis. Although the Notch signalling pathway appears to be a metazoan innovation, Delta-like ligands and Notch-like receptors are present in unicellular relatives of animals, consistent with a proto-Notch pathway playing a role in cell communication

before the evolution of multicellularity. Here we show that Notch is critical for the development of the marine demosponge *Amphimedon queenslandica* as it is necessary for transdifferentiation of epithelial tissues at metamorphosis and juvenile morphogenesis. Notch intracellular domain (NICD) localises to the nuclei of *A. queenslandica* larval epithelial sensory cells and juvenile epithelial choanocytes (feeding cells) during transformation into a mesenchymal stem cell (the archeocyte). In both cases, nuclear translocation of NICD is required for the epithelial-mesenchymal transition and fate conversion in these cells. Nuclear translocation of NICD in the larval sensory cells is induced by a natural environmental cue on the surface of a specific coralline alga and requires actin-driven cell shape changes mediated by calcium signalling. Inhibition of either Notch, calcium signalling or cytoskeletal rearrangements prevent the nuclear localization of NICD and the initiation of metamorphosis. These results support an ancient origin of Notch signalling-dependent cell fate conversion in metazoans, and suggest that environmentally regulated cell fate conversion may have been integral feature of the very first multicellular animals.

C14-04 The evolutionary role of microRNA gene regulation in development: Insights from hemichordates

Gray, Jessica (Harvard Medical School, Boston, MA, USA); Freeman, Jr., Robert (Harvard Medical School, Boston, MA, USA); Gerhart, John (University of California Berkeley, CA, USA); Kirschner, Marc (Harvard Medical School, Boston, MA, USA)

The evolutionary origins of chordate-specific traits remain challenging to explain given the broad conservation of major signaling pathways. One potential explanation of evolutionary change lies in the differential regulation of these conserved pathways during development. The class of short non-coding RNAs known as miRNAs is an intriguing candidate for study of developmental regulatory changes through evolution. miRNAs act as post-transcriptional regulators of gene expression networks, and multiple evolutionary expansions of miRNAs are associated with increasing complexity. However, despite growing data for the importance of miRNAs in the development of model organisms, and genome-wide small RNA studies in numerous species, the functional roles of miRNAs in the development of a wide range of organisms are unknown. The question remains whether the evolution of miRNA targets and functions have driven the evolution of developmental pathways or if they are instead uniquely regulated in different lineages. We are investigating the developmental expression and function of miRNAs in the direct-developing hemichordate *Saccoglossus kowalevskii*. Hemichordates and vertebrates share a common ancestor and many developmental signaling pathways, making it an ideal model for uncovering how ancestral miRNAs may have contributed to the evolution of development in deuterostome lineage. We have used small RNA sequencing to show that *Saccoglossus* miRNAs are dynamically expressed throughout development, suggesting potential roles in a number of developmental processes. In order to identify the function of these miRNAs, spatiotemporal expression data and target predictions are being combined with functional perturbations in the developing embryo. An initial functional screen has confirmed a conserved role for miR-1 in muscle development and ongoing investigations are focused on the regulation of developmental signaling pathways by the neural miRNAs miR-7 and miR-124 in *Saccoglossus*. The targets and functions of *Saccoglossus* miRNAs are being compared with their homologs and functional counterparts in both deuterostomes and protostomes in order to gain insight into the evolving role of miRNAs in development. Our data provide a first exploration of miRNA function in hemichordate development and will contribute to understanding how the role of miRNA regulation in development has changed through evolution.

16.30 – 17.00 Contributed Session C15: EvoDevo as an approach to understanding communication: Modeling, genetics, and developmental research in vocal communication and its neurological underpinnings

ROOM A

Chair: Irene Berra

C15-01 Endless forms of reward. A combinatorial solution for convergent behavioural traits

Berra, Irene (University of Amsterdam, NLD, University of Messina, ITA)

The synthesis between developmental genetics and evolutionary biology can account for both descent and modification of convergent morphological traits, e.g., camera eyes in cephalopods and vertebrates. In a combinatorial solution, the degrees of homology and convergence are specified for any operationally independent level, namely, genetic sequence, expression pattern, neural circuitry, and phenotypic outcome. The traditional distinction between similarity due to common ancestry and apparent similarity due to convergence depends on the focus of analysis. The same holds true for behavioural traits. Broadly shared neuropeptides, i.e., oxytocin, vasopressin, and their fish homologues, have been implicated in a wide array of social and sexual behaviours, such as mating preferences, grooming, nursing, affiliation and its side effect — i.e., out-group competition — across vertebrate species. Even among invertebrates, variations in receptor gene structure can affect the reproductive and social attitudes by reshaping

the receptor distribution of ancestral peptides. This contribution suggests an extension of the combinatorial evo-devo framework to the domains of reciprocity and vocal learning. As for reciprocity, the tendency to conflate ultimate into proximate explanations resulted in easy assumptions of strong cognitive constraints. On the contrary, convergent cooptions of the reward brain system may have lightened the cognitive load required by keeping track of past benefits received. This could be the reason why observational studies on both non-human primates and corvids found evidence of long-term, but not short-term, reciprocity. Similarly, in mammals, the rare capacity to modify the vocal production as a result of experience is socially rewarded by closeness with parents or caretakers. Despite their evolutionary divergence, oscine songbirds and humans independently recruited the same transcription factor and pre-existing brain regions, including the reward system, for the evolution of vocal learning. It is likely that the main differences between speech and birdsong lie in the developmental coordination of genes regulating language. These are just two of the cases in which an evo-devo account of convergence and homology can be applied to our behavioural and psychological classes.

C15-02 Language acquisition as an organic developmental process

Blasco Máñez, Teresa (The KLI Institute, Klosterneuburg, AUT / University of Oviedo, ESP); Lorenzo, Guillermo (University de Oviedo, ESP); Balari, Sergio (Universitat Autònoma de Barcelona, Bellaterra, ESP)

There is a long tradition in biology and psychology to see the cognitive abilities of animals (humans included) as either the product of instinct or the result of some learning process. This dichotomy is deeply entrenched in behavioral sciences such as behavioral ecology, sociobiology and evolutionary psychology (see Laland and Brown 2011 for an overview of these disciplines), causing the analysis of development to be reduced to the mere determination of how much is "genetic" and how much "learned" in some specific cognitive or behavioral activity. The ontogeny of cognition and behavior has thus been neglected from the definitions of "development", which usually place their emphasis on organic structure. Influenced by this tradition, the mainstream assumption within linguistics has been that language acquisition is a proper biological process, even though this assumption has rarely been made explicit beyond the limits of the most naive genetic determinism. A number of authors, however, have seen in EvoDevo the ideal framework for delivering acquisition theory from its biological incoherence by opening the black box of development in order to account for the easiness, speed, and uniformity with which children acquire language (Longa and Lorenzo 2008, 2012; Lorenzo and Longa 2009). In this talk I examine the notion that the acquisition of language, in spite of its dual nature, can be deemed a bona fide organic developmental process (Balari and Lorenzo in press). This idea gets further support from cross-species comparison concerning the capacity of complex vocal learning, once we focus on the deep homology that seems to underlie the emergence and implementation of this capacity (Scharff and Petri 2011) and on the constraints that seem to shape and scaffold the development resulting systems.

16.30 – 17.00 Contributed Session C16: Quo vadis EvoDevo?

ROOM B

Chair: Cassandra Extavour

C16-01 Remaining questions of the developmental hourglass model

Irie, Naoki (University of Tokyo, JPN); Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, JPN)

Do earlier embryos reflect a more ancestral state? In other words, did earlier embryos remain more conservative during evolution? The developmental hourglass model contradicts this idea, predicting that mid-embryonic stages should be the most conservative, and anatomical features of this mid-embryo represent the body plan of each animal phylum. An increasing number of recent molecular-based studies, including ours, support the developmental hourglass model. However, to what extent has the model been proved? In addition to our recent findings based on comparative transcriptome analyses of five vertebrate embryogenesis (mouse, chicken, xenopus, turtle, and zebrafish), I would like to discuss selected, remaining questions toward understanding the general formulation between ontogeny and phylogeny.

C16-02 Arthropod developmental patterns through time: Is there a decline of diversity?

Haug, Joachim (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER); Haug, Carolin (Ludwig Maximilian University of Munich, Planegg-Martinsried, GER)

Many modern arthropod ingroups show rather stereotypic developmental patterns, i.e., within the specific ingroup there is often little to no variation of the ontogenetic pattern. Hence, one would conclude that this type of stereotypic developmental pattern, as shown by all modern representatives of the group, should already be present in the ancestor of the group. We present examples of different groups of reptantian lobsters that do not meet this assumption. One example is achelatan lobsters — spiny lobsters and slipper lobsters. All modern representatives of achelatan lobsters show the same distinct developmental pattern, including a highly specialised giant

larva (the so-called phyllosoma) and a pronounced metamorphosis at the end of the larval phase. Yet, we can show that extinct early representatives of both major lineages in achelatan lobsters, spiny and slipper lobsters, had a less pronounced metamorphosis, i.e., developed more gradually. Hence, the pronounced metamorphosis seen in both modern sub-groups evolved independently and does not represent an ancestral feature of achelatan lobsters. Furthermore, we can show that although today achelatan lobsters possess a single stereotypic larval type, the phyllosoma, early representatives showed a variety of different larval forms. Accordingly, a large diversity of larval forms became lost through time within achelatan lobsters. Similar examples come from other reptantian groups as well as other crustacean lineages for which we can show that now extinct representatives showed a quite different developmental pattern than their modern relatives. We emphasise how important the inclusion of fossil information is for reconstructing ancestral developmental patterns. We finally discuss possible selective pressures that lead to the apparent loss of diversity of developmental patterns within these numerous arthropod lineages.

16.30 – 17.00 Contributed Session C17: Regeneration EvoDevo

ROOM C1

Chair: Uri Frank

C17-01 Distinct mechanisms underlie proximal and distal regeneration in the cnidarian, *Hydractinia echinata*

Bradshaw, Brian (National University of Ireland, Galway, IRL; **Frank, Uri** (National University of Ireland, Galway, IRL)

Cnidarians are renowned for their ability to regenerate any missing body part. Classical work on the freshwater polyp hydra has shown that both foot and head regeneration can occur without a requirement for cell proliferation (i.e., through morphalaxis). This is in contrast with planarians, where stem cell proliferation and blastema formation underlie both head and tail regeneration. Urodele amphibians can regenerate only distally and also require cell proliferation and blastema formation. We have studied both distal (head) and proximal (stolon) regeneration in the colonial marine cnidarian, *Hydractinia echinata*. We show that a burst of stem cell proliferation occurs following decapitation, i.e., during distal regeneration. Some of these cells migrate to the stump and establish a blastema, proliferate and ectopically express not only stemness markers like Piwi, Vasa, Myc and PL-10, but also nematocyte lineage markers like Ncol1, which are required to complete head regeneration. Inhibition of proliferation by pharmacological agents or gamma irradiation inhibited head regeneration. Polyps that have been removed from their colony could also regenerate proximal structures (i.e., stolons) and thereby an entire functional colony de novo. However, stolon regeneration involved a completely different mechanism than head (distal) regeneration. Starting with wound healing, isolated polyps initially did not show any sign of stolon regeneration and fed normally for up to several weeks. Thereafter, they gradually lost anterior-posterior polarity, manifested by loss of tentacles and deregulation of Wnt3 and stem cell marker expression. This was followed by the transformation of the polyp into stolonal tissue that could now attach to the substratum and bud new polyps, establishing a new, fully functional colony, including gamete producing sexual polyps. Head (distal) regeneration can usually take 2 to 3 days to complete while the entire process of stolon, or proximal, regeneration could last for many weeks. Our results show, for the first time, that completely distinct mechanisms of regeneration can act within a single individual to regenerate different body parts.

C17-02 Homoscleromorpha (Porifera) ectosome regeneration: Morphallaxis and metaplasia

Ereskovsky, Alexander (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); Borisenko, Ilya (Saint-Petersburg State University, RUS); Gazave, Eve (Institut Jacques Monod, CNRS, Université Paris-Diderot Paris 7, FRA); Renard, Emmanuelle (Aix-Marseille University, FRA); Borchiellini, Carole (Aix-Marseille University, FRA)

The ability to regenerate is widespread into the animal kingdom. Sponges are known to possess remarkable reconstitutive and regenerative abilities ranging from the common wounding or body part regeneration to the more impressive re-building of a functional body from dissociated cells. Among the four sponge classes, Homoscleromorpha is notably the only sponge group to possess morphologically distinct basement membrane and specialized cell junctions, thus possessing true epithelia. The consequence of this peculiar organization is the predominance of epithelial morphogenesis during development, metamorphosis, and asexual reproduction of these sponges. The objective of this work is to reveal the underlying cellular mechanisms used during morphogenesis accompanying ectosome regeneration in our homoscleromorph sponge model: *Oscarella lobularis*. During regeneration of exopinacoderm and restoration of functional peripheral parts of aquiferous system in *O. lobularis* three main sources of new exopinacoderm have been evidenced by microscopy electronic: (1) intact exopinacoderm, surrounding the wound surface, (2) the layers of endopinacoderm of peripheral exhalant and inhalant canals, and (3) the intact choanoderm find on the wound surface. The basic morphogenetic processes during regeneration are spreading and fusion of epithelial sheets (exo- and endopinacoderm) that merge into one continuous

epithelium. Transdifferentiation of choanocytes and apopylar cells into exopinacocytes are also present. In *O. lobularis*, the archaeocytes (totipotent cells of demosponges) are absent. Moreover, we cannot reveal any other morphologically distinct totipotent cells. Any elements of epithelial-mesenchymal transition are also absent during regeneration. The number of DNA-synthesizing cells and their distribution in the tissue was not appreciablychanged during regeneration, as shown by EdU. The regeneration in *O. lobularis* passes through cells transdifferentiation and through morphallaxis, when lost body parts are replaced by the remodeling of the remaining tissue. Morphogenesis during ectosome regeneration in *O. lobularis* is correlated with its true epithelial organization. Knowledge of morphological basis of morphogenesis during *Oscarella* regeneration could have important implications for our understanding of the diversity and evolution of regeneration mechanisms in metazoans.

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16.30 – 17.00 **Contributed Session C18:**
Ecological and environmental impacts on the evolution of organismal development II

ROOM C2

Chair: Angelika Stollewerk

C18-01 Regulatory gene networks that shape the development of adaptive phenotypic plasticity in a cichlid fish

Schneider, Ralf Friedrich (University of Konstanz, GER); Li, Yuanhao (University of Konstanz, GER); Meyer, Axel (University of Konstanz, GER); Gunter, Helen M (University of Konstanz, GER)

The ability of a genotype to produce variable adaptive phenotypes according to environmental stimuli is termed “phenotypic plasticity”, a factor that may have driven major adaptive radiation events. Despite its ecological and evolutionary importance, the developmental regulatory networks underlying developmental plasticity remain largely uncharacterised. The cichlid fish *Astatoreochromis alluaudi* exhibits pronounced plasticity in response to a mechanically stimulating diet in the pharyngeal jaw apparatus, a key innovation that is believed to have promoted the spectacular diversification observed amongst East African cichlids. We gained insight into the regulatory basis of this plasticity by examining developmental expression of 19 previously identified “plasticity genes” in specimens that were raised for between one and eight months on either a hard or soft diet (whole snails or finely pulverised snails respectively). Plastic morphologies were first detected after three to five months of treatment. Interestingly, differential expression of our candidate plasticity genes preceded the onset of observable morphological divergence, suggesting that their expression contributed to the initiation of the plastic phenotypes. Striking co-expression was observed amongst our candidates that belong to similar functional classes, suggesting that these genes display modular patterns of regulation and acting in concert to orchestrate the development of this adaptive trait. To investigate the regulatory basis of this modular expression, we conducted a transcription factor binding site analysis. Our results allowed us to construct a regulatory network that underlies the observed plasticity. The mechanically responsive transcription factor AP1 was identified as a putative major regulator of the plastic response, influencing gene expression on various hierarchical levels. By investigating phenotypic plasticity throughout a developmental time-course we have identified an environmentally responsive, interconnected regulatory network that underlies the development of the integrated plastic LPJ phenotypes in a recently established molecular model for adaptive plasticity.

C18-02 Running for life: Developmental and biomechanical constraints on homeotic transformations in mammals

Galis, Frietson (Naturalis Biodiversity Center, Leiden, NLD); Carrier, David (University of Utah, Salt Lake City, UT, USA); van Alphen, Joris (Groningen University, NLD); Metz, Johan (IIASA, Laxenburg, AUT); Ten Broek, Clara (Naturalis Biodiversity Center, Leiden, NLD)

The mammalian vertebral column is highly variable, reflecting adaptations to a wide range of lifestyles, from burrowing in moles to flying in bats. Yet, in many taxa the number of trunk vertebrae is surprisingly constant. We argue that the latter constancy results from strong selection against initial changes of these numbers in fast-running or agile mammals, while such selection is weak in slower-running, sturdier mammals. The rationale is that changes of the number of trunk vertebrae require homeotic transformations from trunk into sacral vertebrae, or vice versa, and mutations towards such transformations generally produce transitional lumbosacral vertebrae that are incompletely fused to the sacrum. We hypothesize that such incomplete homeotic transformations impair flexibility of the lumbosacral joint and, thereby threaten survival in species that depend on axial mobility for speed and agility. Such transformations will only marginally affect performance in slow sturdy species, so that sufficient individuals with transitional vertebrae survive to allow eventual evolutionary changes of trunk vertebral numbers. We present data on fast and slow carnivores and artiodactyls and on slow afrotherians and monotremes that strongly support this hypothesis. The conclusion is that the selective constraints on the number of trunk vertebrae stem from a combination of developmental and biomechanical constraints.

16.30 – 17.00 Contributed Session C19:
Amphioxus EvoDevo

ROOM D

Chair: Beatriz Albuixech-Crespo

C19-01 Molecular patterning of amphioxus CNS reveals unexpected evolutionary relationships between midbrain and diencephalon

Albuixech-Crespo, Beatriz (University of Barcelona, ESP); Irimia, Manuel (University of Barcelona, ESP); Maeso, Ignacio (University of Barcelona, ESP); Sánchez-Arrones, Luisa (CSIC-UAM, Madrid, ESP); Somorjai, Ildiko (University of St Andrews, GBR); Pascual-Anaya, Juan (Riken Center for Developmental Biology, Kobe, JPN); Bovolenta, Paola (CSIC-UAM, Madrid, ESP); Garcia-Fernàndez, Jordi (University of Barcelona, ESP); Ferran, José Luis (University of Murcia, ESP); Puelles, Luis (University of Murcia, ESP)

The vertebrate central nervous system (CNS) is built following a common, highly conserved construction plan — bauplan — that is defined by basic anterior-posterior and dorsoventral subdivisions. These partitions, or neuromers, are characterized by differential expression of specific gene markers. The conservation of the neuromers and their associated genoarchitectonic patterns in all vertebrate groups indicates the presence of a deeply conserved anatomical and molecular bauplan of the CNS in the last common ancestor of vertebrates. However, it is still unclear to what extent this bauplan is conserved. We studied the expression of ~50 genes with key roles in vertebrate neural patterning in amphioxus that, unlike non-chordates, develops a neural plate that is homologous to that of vertebrates. These molecular and topological data are compiled in the most comprehensive model of molecular regionalization of the incipient neural tube of the cephalochordate amphioxus, the most basal-branching chordate. Comparison of topologically adjusted molecular data between amphioxus and vertebrates suggests that amphioxus presents a simplified chordate bauplan; however, its basic partitions seem to have clear homology and common ancestry with those of vertebrates showing unexpected developmental and evolutionary relationships between well-established vertebrate encephalic regions.

C19-02 Functional study of neural induction in the cephalochordate, *Branchiostoma lanceolatum*

Le Pétillon, Yann (UPMC – Laboratoire Arago, Banyuls-sur-Mer, FRA)

Neural induction is the process through which pluripotent cells are committed to a neural fate. This mechanism is controlled in the vertebrate embryo by a peculiar region called the organizer. A quite widely accepted model states that in vertebrates both BMP signal inhibition and an instructive signal, that may be FGF, are required in ectodermal cells to induce neural fate acquisition. On the other hand, in tunicates, which represent the sister group of vertebrates, BMP signaling pathway is not implicated in neural induction which is exclusively under the control of the FGF signal. However, tunicates have lost the organizer which might explain this divergence compared to vertebrates, and the question of how neural induction was controlled in the ancestor of chordates remains to be addressed. Here we show that the cephalochordate amphioxus, which represents the third group of chordates, possesses an organizer able to trigger neural induction and that this process is controlled by BMP signal inhibition together with activation of Activin/Nodal and/or FGF signaling pathways. Using classical embryology methods, we demonstrate that the dorsal blastoporal lip of the amphioxus gastrula has the same properties as the vertebrate organizer. We also show that BMP is the signal triggering epidermal program entry and that its inhibition is necessary but not sufficient for neural induction. Moreover, using ectodermal explants, we observe that FGF or Nodal/Activin signals are able to induce the entry of ectodermal cells in the neural program and that these pathways act independently of each other. Altogether, our data suggest that neural induction in the ancestor of chordates was controlled by an organizer embryonic structure through BMP inhibition and activation of another pathway, probably FGF and/or Activin/Nodal. Although Activin/Nodal pathway is thought to act mainly as an inhibitor of neural induction in vertebrates, our results strongly plead for a deeper examination of the implication of this pathway in the first step of nervous system formation. Our evolutionary approach opens new avenues for a deeper comprehension of the fine molecular events controlling the complex developmental process of neural induction.

17.10 – 17.20 Kowalevsky Medal to
Denis Duboule

ROOMS C1&2 (University of Geneva, CHE)

17.20 – 18.00 Keynote Lecture (K3)
The evolution of key bilaterian traits: Insights from regulatory developmental networks in Cnidaria

ROOMS C1&2

Ulrich Technau
(University of Vienna, AUT)
Chair: Michael Schubert

Cnidaria, the sister phylum of Bilateria, lacks a number of key bilaterian traits, such as mesoderm, a central nervous system and a clear bilaterality, yet the underlying genetic basis for these crucial differences is unknown. Genome sequencing projects have revealed that the anthozoan Nematostella vectensis displays a stunning ancestral complexity in gene repertoire, gene structure and genome

organisation. Therefore, differences in the cis-transcriptional or post-transcriptional regulation as well as protein interactions may account for differences in functions of conserved genes. To this end, we mapped cis-regulatory elements (promoters and enhancers) on a genome-wide level using a combination of histone modifications and binding of Pol II and of transcriptional cofactor p300. We also analysed the repertoire and the mode of action of miRNAs in post-transcriptional regulation. Lastly, I will report that complex signaling networks with conserved components and non-conserved modulators have possibly evolved independently in Cnidaria and Bilateria to pattern a secondary body axis.

18.00 – 20.00 **Workshop:**
Advances in live imaging morphogenesis

ROOM A [& ROOM F]

Chairs: Frederike Alwes and Carsten Wolff
Demonstration

W-01 High-speed light sheet microscopy and real-time image processing

Huisken, Jan (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

Light sheet microscopy, such as Selective Plane Illumination Microscopy (SPIM), reduces phototoxic effects to a minimum. Due to the illumination of the sample in a thin volume around the focal plane no tissue outside the plane of interest is exposed and bleached. In addition, the fluorescence is collected with highly sensitive cameras. Combined with novel sample mounting techniques, SPIM has become a powerful technique for the long-term observation of fragile biological organisms. SPIM benefits from the latest camera technology and is therefore constantly improving in speed and sensitivity. Over the years it has become evident that light sheet microscopy is revolutionizing bioimaging in several ways. Lately, we have shown that three-dimensional (3D) volumes can be imaged almost instantaneously using electrically tunable lenses (ETL) in SPIM. This makes SPIM the fastest fluorescence microscopy technology for non-invasive 3D imaging. Even the dynamics of the beating zebrafish heart can be captured and the myo- and endocardial tissues as well as the blood can be visualized by 3D reconstruction. Experiments have become possible that run at full speed using the best possible hardware without being limited by the fragility of the sample. The speed advantage of the SPIM over other fluorescence technique can be utilized not only to image rapid events in developing tissues but also to record a large number of views for multi-view reconstruction. One key application of light-sheet technology includes the multi-dimensional imaging of the developing zebrafish larvae over extended periods of time. I will give some examples of the unique capabilities of SPIM, especially for monitoring the development of the zebrafish heart and the early endoderm.

W-02 SPIM imaging of spiralian development

Handberg-Thorsager, Mette (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Tomer, Raju (Stanford University, CA, USA); Amat, Fernando (Janelia Farm Research Campus, Ashburn, VA, USA); Girstmair, Johannes (University College London, GBR); Telford, Max (University College London, GBR); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER); Keller, Philipp (Janelia Farm Research Campus, Ashburn, VA, USA); Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

The development of light-sheet microscopy for the analysis of biological processes is a landmark in biology. Compared to standard fluorescent imaging techniques, light sheet microscopy offers lower phototoxicity and photobleaching of the fluorophores and the ability to image large specimen from multiple sides resulting in better coverage. Light-sheet microscopy achieves this through the combination of illuminating only the plane in focus (i.e., the imaged plane), with, in some cases, the use of more than one illumination objective and more than one detection objective or imaging of the specimen from multiple angles. In combination with image processing techniques, light sheet microscopy has the potential to follow entire embryos with cellular resolution throughout development. This paradigm shift in microscopy will also mark a new era in EvoDevo, because with only a few tools, including a functional injection protocol, one can address many biological questions in less-established model organisms. We use light-sheet microscopy to follow the early developmental events of lophotrochozoans including the marine annelid polychaete, *Platynereis dumerilii* (with the Keller and Arendt labs) and the marine polyclad flatworm, *Maritigrella crozieri* (with the Telford lab). We are interested in studying the common principles of the spiralians given that the spiralian cleavage mode is ancestral for the lophotrochozoans. In this talk I will focus mainly on our data from *P. dumerilii* and present some of the developmental events that we can now visualize by means of light-sheet microscopy. I will demonstrate an automated tracking tool for cell lineaging (by F. Amat and P. Keller) and a visualization program CATMAID (Saalfeld et al. 2009) for the visualization of the segmented and tracked cells.

W-03 Imaging and quantifying 4D growth patterns during flower development

Das, Pradeep (ENS Lyon, Lyon, FRA)

The emergence of stereotypical shapes in tissues and organs requires the coordinated regulation of specific growth patterns across space and time during development. We seek to gain a clear understanding of how the underlying molecular, genetic and physical events govern growth in the Arabidopsis flower. The most obvious way to measure and describe growth is at the cellular level. Over the last several years, we have developed a software pipeline to computationally track the growth of Arabidopsis flowers at cell resolution and in 4 dimensions (space and time). We are now in the process of using segmented time course data to statistically analyse floral growth leading up to the first morphogenetic events. Naturally, these patterns are a result of the genetic and mechanical events occurring during development. To this end, we are also examining the link between growth and patterning, and between patterning and mechanics. I will describe the challenges we face in generating and treating our confocal imaging-based 4D floral reconstructions. I will then describe the analysis toolkit we are developing to uncover cellular growth metrics, which should be applicable to 4D data obtained from any other imaging setup (light sheet microscopy, SPIM etc.) and processed with any software (Openalea, MorphographX etc.).

W-04 Multi-level studies of appendage morphogenesis in the crustacean model, *Parhyale hawaiensis*

Pavlopoulos, Anastasios (Howard Hughes Medical Institute, Ashburn, VA, USA); Tinevez, Jean-Yves (Institut Pasteur, Paris, FRA); Pietzsch, Tobias (Max Planck Institute, Dresden, GER); Wolff, Carsten (Humboldt-Universität, Berlin, GER); Tomancak, Pavel (Max Planck Institute, Dresden, GER)

We are interested in the molecular, cellular and mechanical control of tissue and organ morphogenesis during animal development. The study of morphogenesis requires capturing and integrating information across multiple levels of biological organization: from the subcellular level to single cell behaviors, to emerging biological forms. In order to bridge these scales, we have focused on arthropod models that satisfy a number of appealing biological and technical requirements. I will describe our studies on the crustacean model *Parhyale hawaiensis* that offers several advantages to follow appendage morphogenesis from early specification until late differentiation stages. *Parhyale* embryos have the right optical properties for microscopic live imaging at a very high spatial and temporal resolution with multi-view fluorescence light-sheet microscopy. To stand up to the challenges these multi-view terabyte-sized recordings raise, we are continuously developing the software needed for image processing and image analysis that is free and easy to use through the Fiji bioimaging platform. With these tools in hand, we can reconstruct the cell lineages and quantify a number of cell behaviors underlying morphogenesis and diversification of neighboring serially homologous appendages in developing *Parhyale* embryos. *Parhyale* research is also supported by an increasing number of functional genetic approaches, embryological manipulations, genomic and transcriptomic resources. We are taking a number of unbiased and candidate gene approaches to visualize and manipulate gene expression and function *in vivo* and make the link between gene activity and morphogenetic cell behaviors. This research line aims to uncover the fundamental principles governing the self-organization and function of living systems, from the sub-microscopic to the macroscopic level.

W-05 Light sheet microscopy as a tool for studying early insect development

Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

Light sheet fluorescence microscopy has emerged in recent years as an excellent tool for studying dynamic developmental processes. Compared to classical confocal techniques, light sheet microscopy is gentler to the imaged specimen, allowing long term imaging in three dimensions. Particularly, the Selective Plane Illumination Microscopy (SPIM), a flavour of light sheet technology that involves sample rotation, enables imaging of relatively large specimen in their entirety. Taken together, light sheet microscopy makes it possible to record developmental anatomy of a non-model organism embryo and achieve in one experiment the level of description of developmental anatomy that would take decades of painstaking research before. This power comes for a price, since the technology is new, rapidly growing and requires quite substantial support from computer science — a scientific discipline that many biologists are less familiar with. I will discuss our efforts to bring the light sheet microscopy technique into the hands of practicing biologists and to make the associated image processing as straightforward as possible. We use open access principles both for software and hardware to reach these goals. I will present how these tools enable us, and hopefully others, to study early developmental morphogenesis in various model systems with ease and unprecedented resolution.

18.00 – 20.00
Corridors C and ROOM E

Poster Session 2
(odd numbers)

Friday, July 25th

09.00 – 10.40 **Symposium S18:**
EvoDevo of cranial neural crest populations across developmental systems

ROOM A

Organizer: Georgy Koentges
Chair: Georgy Koentges

S18-01 Characterizing evolutionary conserved regulatory networks in zebrafish craniofacial development

Eberhart, Johann (University of Texas at Austin, TX, USA); Swartz, M. E. (University of Texas at Austin, TX, USA); Wells, M. B. (University of Texas at Austin, TX, USA); Li, Q. (University of Texas at Austin, TX, USA); Sheehan-Rooney, Kelly (University of Texas at Austin, TX, USA); Rozacky, Jenna (University of Texas at Austin, TX, USA); Dixon, M. J. (University of Manchester, GBR); Vokes, Steven A. (University of Texas at Austin, TX, USA)

We are interested in the genetic hierarchies that regulate cell behaviors during craniofacial morphogenesis. We have found that zebrafish have a striking similarity in the expression and function of genes involved in amniote craniofacial development. For instance, *Satb2* is expressed in the maxillary and mandibular mesenchyme in mouse, chicken and zebrafish. Using zebrafish, we have found that the proper expression of *satb2* depends upon both Bmp and Shh signaling to the mesenchyme. To better characterize the regulation of *satb2*, we have begun to examine the *cis*-regulatory modules driving expression in the craniofacial mesenchyme. In a mouse Gli-binding region dataset, we found a 974 bp region upstream of *satb2* that drives robust expression in the mandibular mesenchyme of mouse G0 embryos and zebrafish larvae. By searching for evolutionary conserved sequences within this Gli-binding region, we identified a 100 bp sequence that is conserved from human to the lobe-finned fish, coelacanth. We have found that both the mouse and coelacanth sequences recapitulate the mandibular enhancer activity of the full-length Gli-binding region. We are currently characterizing the involvement of conserved predicted transcription factor binding elements within this enhancer. These results will shed light on the conservation of craniofacial development across vertebrate species.

S18-02 Variation in craniofacial derivation and development: Insights from extreme model systems

Gross, Joshua (University of Cincinnati, OH, USA)

The neural crest has long been appreciated as the direct cellular source for much of the vertebrate craniofacial skeleton. Historical fate-mapping studies, stretching back to the early 20th century, revealed a pattern of highly conserved neural crest contributions across taxa. Recent studies, however, demonstrate significant variation in the precise derivatives of the adult skull in a common anuran system, *Xenopus laevis*. The developmental origin of this variation remains unclear, however it may be a consequence of extreme life history changes associated metamorphosis that must occur prior to skull ossification in these organisms. Another emerging model system, *Astyanax mexicanus*, similarly demonstrates significant changes to the craniofacial complex as a consequence of life in the extreme, subterranean habitat. Following >1My in total darkness, cave dwelling forms evolved numerous alterations to the craniofacial skeleton under genetic control. One trait in particular, dermal bone fragmentation of the neural crest-derived facial skeleton, is frequently observed on one side of the skull, but not the other, in cave dwelling forms. QTL studies revealed an asymmetric genetic basis for this cave-associated trait, however the embryonic origins of these aberrations remain unknown. Interestingly, surface dwelling forms exhibit perfectly symmetric craniofacial features. Given the persistence of cave and surface dwelling forms, this extreme model system will enable powerful novel insights to the molecular and developmental underpinnings of aberrant craniofacial asymmetry.

Supported by NIH/NIDCR DE022403.

S18-03 Elaborating a forebrain: Role of the neural crest in vertebrate evolution

Creuzet, Sophie (Institut de Neurobiologie Alfred Fessard, Gif-sur-Yvette, FRA)

Encephalization is the most important characteristic in the evolutionary transition leading from protochordates to vertebrates. This event has coincided with the emergence of a transient and pluripotent structure, the neural crest (NC), which is absent in protochordates. In vertebrates, NC provides the rostral cephalic vesicles with skeletal protection and functional vasculature. Our investigations show that, aside from its structural role in craniofacial ontogenesis, the NC exerts a potent morphogenetic "paracrine" role on the brain and sense organs development. Understanding how the NC conveys its trophic effect is the focus of our research. Our work opens up new avenues for understanding the molecular mechanism whereby the NC has accompanied the development of the forebrain. Our data provide an explanation of the fact that the advent of the NC has enabled the elaboration of the cephalic alar plate and the considerable expansion of telencephalic hemispheres.

S18-04 Comparative rhombomeric fate mapping of neural crest and its evolutionary implications

Koentges, Georgy (University of Warwick, Coventry, GBR)

Neural crest cell populations are at the heart of craniofacial morphology and evolution. We have recently started with colleagues to perform comparative fate mapping of identical populations across different taxa using genetic and single cell transplantation approaches, in order to tease out similarities and differences that might be instrumental in the key changes of craniofacial shape. I will be talking about our first results in this endeavour and their evolutionary implications for macroevolutionary transitions.

09.00 – 10.40 Symposium S19: Quantitative EvoDevo in model and non-model organisms I

ROOM B

Organizers: Benedikt Hallgrimsson, Chris Klingenberg, Philipp Mitteroecker and Ruth Flatscher
Chair: Christian Klingenberg

S19-01 Morphometrics and the developmental genomics of canalization in craniofacial development

Hallgrimsson, Benedikt (University of Calgary, AB, CAN); Gonzalez, Paula M. (Universidad Nacional de La Plata, ARG); Mio, Washington (Florida State University, FL, USA); Young, Nathan M. (University of California San Francisco, CA, USA); Percival, Christopher (University of Calgary, AB, CAN); Liberton, Denise (Pennsylvania State University, University Park, PA, USA); Jamniczky, Heather (University of Calgary, AB, CAN); Marcucio, Ralph (University of California San Francisco, CA, USA)

Canalization refers to the suppression of either genetic or environmental variance. The evolution of mechanisms that regulate the expression of variance is a fundamental feature of complex developmental systems. The genetic and developmental basis for this phenomenon, however, is largely unknown. There are two basic views on this issue. In one, there are specific evolved mechanisms such as heat shock proteins that suppress or regulate the expression of variance in response to environmental or genetic perturbations. In the other, canalization occurs as an emergent property of developmental systems. We review our recent work on the developmental genetics of variation in vertebrate craniofacial morphology in avian and mouse models and a genomic approach to human facial shape variation. We argue that the evidence from these studies overwhelmingly supports the latter view. Canalization effects emerge from nonlinearities in development, from changes to patterns of network redundancy and from dysregulation of morphogenetic processes. Non-linearities emerge at multiple levels in development. We illustrate the use of novel morphometric approaches to investigate this phenomenon during face formation. In particular, we hypothesize that the 3D morphology of gene expression contributes to nonlinear relationships between pathway activation and craniofacial shape. The genomics of phenotypic variance for human facial shape supports the emergent property view of canalization as well. Genetic effects on variance appear to be small and widely distributed across the genes that influence craniofacial shape. This work illustrates the utility of embedding a morphometric approach in developmental and genomic approaches to complex morphological traits.

S19-02 An information theoretic approach to identifying cranial modularity with 3-D morphometric data

Goswami, Anjali (University College London, GBR); Finarelli, John (University College Dublin, Dublin)

Identification of modules can use exploratory approaches (e.g., cluster analysis) or confirmatory ones (e.g., RV coefficient analysis). Confirmatory approaches are more robust, but most suffer from an inability to compare models with different parameterizations. For example, both a two-module neurocranial/facial model and a more complex six-module model may be significantly supported for the therian mammalian skull using RV coefficient analysis. Here we present an approach to analyze cranial modules with 3-D geometric morphometric data that takes model parameterization into account. We employed a dataset of 61 landmarks from 181 macaque skulls (*Macaca fuscata*), split into five groups (deciduous teeth only, erupted M1, erupted M2, adult females, adult males). We generated model log-likelihoods of trait correlation matrices, which were compared with the finite-sample corrected Akaike Information Criterion. We tested 27 models, including no modules, and various partitions of 2, 3, 4, 6, and 8 modules, based on existing hypotheses of skull development. Our results clearly supported a complex 6-module model, with separate within- and inter-module correlations as the best fit for the adult datasets. No other models are within 20 LnL of this model, but for most datasets the top alternative models are less parameterized variations on the same pattern, for example with six modules but shared distributions for all inter-module correlations. In addition, this pattern of integration appears early in development, with identical models supported for all stages from infants to adults. These results thus support a complex model of modularity for the macaque skull and suggest that this pattern is conserved through late ontogeny.

S19-03 Streptophyta): Quantitative morphometrics at the cellular level

Neustupa, Jiri (Charles University in Prague, CZE)

Morphological symmetry of cellular shapes is an omnipresent feature in protists and different types of complex symmetry are particularly exhibited in freshwater microalgae, such as the desmids (Desmidiales). Their cells are typically composed of two bilaterally symmetric semicells arranged according to two perpendicular axes of symmetry. New semicells develop after mitotic division so that each cell is composed of two unequally old, bilaterally symmetric parts. The peculiar morphological patterns of the desmids were used for addressing several questions regarding their environmentally induced plasticity and shape allometry. Mapping morphological diversification of the Micrasterias lineage on the molecular phylogenetic trees revealed an accelerated rate of morphological evolution, including acquisition or loss of the complex symmetric parts. Plasticity and development of the symmetric structures of Micrasterias cells were found to be associated with key environmental factors, such as temperature or pH. In addition, positive allometric scaling of cell volumes and surfaces revealed that evolution of the intricate morphological patterns of the desmid cells can be related to the selection pressure for maximizing the exchange surface areas.

S19-04 Developmental canalization in the vertebrate cranium: A morphometric approach

Mitteroecker, Philipp (University of Vienna, AUT)

Canalization against internal and environmental fluctuations is a key property of non-pathological development. The underlying mechanisms and molecular components, however, are still poorly understood. I present one formalization of developmental canalization, which allows for the statistical mapping of the strength and pattern of canalization throughout an investigated time period. C. H. Waddington's metaphor of the epigenetic landscape thus turns into an actual estimable statistical property of development. By applications to the vertebrate cranium, I show how different cranial shape features differ fundamentally in the generation and canalization of developmental variance. Functionally relevant features appear to be more tightly canalized than other traits.

09.00 – 10.40 Symposium S20:
Less is more: Loss of gene functions as a driving force of developmental evolution

ROOM C1

Organizers: Cristian Cañestro and Ingo Braasch
Chairs: Cristian Cañestro and Ingo Braasch

S20-01 Dynamic gain and loss of genes in animal evolution

Holland, Peter (University of Oxford, GBR); Quah, Shan (University of Oxford, GBR); Maeso, Ignacio (University of Oxford, GBR); Marletaz, Ferdinand (University of Oxford, GBR); Paps, Jordi (University of Oxford, GBR); Olson, Peter (Natural History Museum, London, GBR); Martin, Kyle (University of Oxford, GBR); Hui, Jerome (The Chinese University of Hong Kong, CHN)

Gene duplication and gene loss are complementary processes. Using examples from mammals, insects and other animals, including homeobox and miRNA genes, I will discuss how gain of genes can occur through a dynamic evolutionary process, balanced by equally rapid gene loss. The net result of such balance would be stasis. In rare cases, new genes get incorporated more deeply into developmental pathways and subsequently less prone to loss. However, on occasion even conserved and ancient genes will be lost, presumably in association with restructuring of developmental pathways.

S20-02 Ohnologs: Why do basal bony fish hold'em but crown groups fold'em?

Braasch, Ingo (University of Oregon, Eugene, OR, USA); Batzel, Peter (University of Oregon, Eugene, OR, USA); Amores, Angel (University of Oregon, Eugene, OR, USA); Ferrara, Allyse (Nicholls State University, Thibodaux, LA, USA); Fontenot, Quenton (Nicholls State University, Thibodaux, LA, USA); Bobe, Julien (Laboratoire de Physiologie et génomique des poissons, INRA, Rennes, FRA); **Postlethwait, John** (University of Oregon, Eugene, OR, USA); Guiguen, Yann (Laboratoire de Physiologie et génomique des poissons, INRA, Rennes, FRA)

Multiple paralogs in vertebrate genomes likely arose from whole genome duplication (WGD) events at the base of the vertebrate radiation. Some have thought that these genome expansion events provided opportunities for variation in genetic information that allowed vertebrates to invent themselves. Recent genome sequencing of coelacanth (a basally diverging lobefin fish), and spotted gar (a basally diverging rayfin fish), shows that these ancient lineages retain a fuller set of ohnologs (i.e., WGD-derived paralogs) than their respective crown groups, the tetrapods and the teleosts. This raises the hypothesis that selective ohnolog loss is associated with innovations derived in crown group evolution. Informed by the coelacanth and spotted gar genomes, we will explore the types of ohnologs gone missing from tetrapods, from teleosts, and from both taxa to help understand how less is more.

S20-03 Revealing cryptic pan-vertebrate gene repertoire in developmental phylome

Kuraku, Shigehiro (RIKEN Center for Developmental Biology, Kobe, JPN)

Conservation of "toolkit genes" has been one of the key concepts in Evo-Devo, but this premise, which prevailed in the pre-genomic era, should now be revisited. Employing genome-wide resources, I and my collaborators have performed intensive molecular phylogenetic analyses and identified multiple developmental regulatory genes, including *Bmp16, Pax4, Pax10* and *Hox14*, which existed in the ancestral vertebrate genomes but were secondarily lost later in multiple lineages ("cryptic pan-vertebrate genes") . Our investigation encompassed the effect of gene loss in our understanding of so-called "two-round (2R)" whole genome duplications and jawless fish genomes as well as dynamics of developmental gene repertoire between different amniote lineages. Overall, not only gene gain but also its loss has created diversity in gene repertoire, which should be addressed carefully before asking whether protein-coding or *cis*-regulatory changes of conserved "toolkit genes" contributed to evolution.

S20-04 A systematic approach to identify gene losses in genome alignments

Hiller, Michael (Max Planck Institute for Molecular Cell Biology and Genetics (MPI CBG) & Max Planck Institute for the Physics of Complex Systems (MPI PKS), Dresden, GER); Sharma, Virag (MPI CBG & MPI PKS, Dresden, GER); Langer, Bjoern (MPI CBG & MPI PKS, Dresden, GER); Foerster, Leo (MPI CBG & MPI PKS, Dresden, GER); Kiruvale, Pradeep (MPI CBG & MPI PKS, Dresden, GER)

A key question in evolutionary biology is what are the changes in the genome that are involved in phenotypic differences between species. Losses of ancestral genes are one type of genomic change with likely functional consequences. Such gene losses have been associated both with the loss of ancestral phenotypes as well as phenotypic novelty. To better understand how gene losses contribute to phenotypic change between species, it would be desirable to identify gene losses in many sequenced genomes in an automated fashion. However, at the level of different species, this is a challenging problem and studies are typically limited to a small number of selected candidate genes and require extensive manual curation. We describe a computational pipeline that we have developed to systematically search for losses of coding genes just using the gene annotation of reference genome and genome alignments of other species. Our pipeline is able to identify the different types of gene inactivating mutations such as stop codons, frame shifts and splice site mutations, while controlling for artifacts that mimic gene loss such as genome assembly issues, alignment ambiguities and gene structure changes. We applied our approach to available mammalian genomes and assess sensitivity and specificity by using known gene inactivating mutations from literature and the "core eukaryotic genes" (highly conserved genes that are used to assess the completeness of new genome assemblies). The design of our pipeline is general and it can be applied to genome alignments of other species and other clades. Compiling gene-loss catalogues for available and future genomes will be one important step towards better understanding how gene losses contribute to phenotypic change.

09.00 – 10.40 Symposium S21: EcoEvoDevo: Symbiosis and epigenetic inheritance

ROOM C2

Organizers: Scott Gilbert and Yoav Soen
Chairs: Yoav Soen and Scott Gilbert

S21-01 EvoDevo of the holobiont

Gilbert, Scott (University of Helsinki, FIN)

The holobiont is the organism with its persistent symbionts. A coral, with its symbiotic algae, and a cow, with its rumen bacteria, are classical examples of holobionts. Coral cannot survive without the carbon resources and oxygen provided by the symbionts, and the cow cannot digest grass without the symbiotic community in its rumen. However, recent research has found that each animal probably develops as a community of organisms, and many signals for organ formation have been "outsourced" to the symbionts. So if we develop through symbiotic relations, then evolutionary developmental biology has to be one an evo-devo of holobionts. Data will be reviewed showing that there are several ways that symbionts contribute to evolution: (1) as a source of phenotypic variation; (2) as providing mechanisms of speciation; and (3) as a source of novelty, potentiating new transitions. In each case, developmental regulation is effected.

S21-02 Experimental evolution of legume endosymbionts

Masson, Catherine (Laboratory of Plant-Microbe Interactions, INRA, Toulouse, FRA)

Rhizobia are phylogenetically dispersed bacteria that have evolved the environmentally essential function of fixing atmospheric nitrogen in symbiosis with legumes. Ample evidence indicates that horizontal transfer of symbiotic genes has played a crucial role in rhizobia evolution. Yet, adaptive mechanisms that allow the recipient genomes to express symbiotic traits are unknown. We experimentally evolved a soil bacterium into legume symbionts using a "design then evolve" strategy. We transferred the symbiotic plasmid of the *Mimosa* symbiont *Cupriavidus taiwanensis* into the phytopathogen *Ralstonia solanacearum*, generating a non-nodulating chimera that was subsequently evolved using serial plant-bacteria co-cultures (Marchetti et al., PLoS Biol 210; Guan et al., ISME J. 2013). Evolution was surprisingly fast since the first

two major symbiotic steps, induction of root nodules and intracellular infection of nodule cells, were not only activated but also dramatically improved over 17 cycles (~400 generations). Adaptive mutations crucial for the transition from pathogenicity towards mutualism targeted the virulence pathway of R. solanacearum. Our findings predict that natural selection of adaptive changes following horizontal transfer has been a major driving force in rhizobia evolution. Moreover we provide evidence for a mechanism that may have facilitated post-HGT adaptation of emerging rhizobia to their new host.

S21-03 Transgenerational inheritance of small RNAs in *C. elegans*

Rechavi, Oded (Tel Aviv University, ISR)

The inheritance of acquired characteristics is a topic of long-standing interest and controversy. While some of the classic Lamarckian ideas have been dismissed, more recent studies suggest that certain traits acquired by an animal during its lifetime may be transmitted to next generations. *C. elegans* inherits acquired antiviral and transposon resistance (in a "Lamarckian" fashion) through transgenerational transmission of small RNAs, which mediate RNA interference. Inherited small RNAs, which protect the worm, can be passed down to many ensuing generations in a non-Mendelian manner, and "vaccinate" RNAi-deficient progeny. Most genes are regulated by different endogenous regulatory small RNA species, and therefore small RNA inheritance might affect the inheritance of many traits. Our recent efforts suggest that inherited small RNAs may indeed reflect the ancestral environment, and possibly prepare the progeny for relevant hardships.

S21-04 Linking EcoDevo with EvoDevo by non-mendelian inheritance of epigenetic and symbiotic changes

Soen, Yoav (Weizmann Institute of Science, Rehovot, ISR)

The development of organisms must be robust enough to maintain adaptive patterns and flexible enough to cope with fluctuating environmental, epigenetic and microbial conditions. How this flexibility is achieved and whether and how it is connected to longer-term establishment of new adaptations are not clear. We are addressing these questions by studying stress-induced induction and inheritance of altered developmental patterns in flies. We identified epigenetic and microbiome-mediated mechanisms that promote increased developmental flexibility under stress and contribute to non-Mendelian transfer of influences across generations. I will discuss these mechanisms and their potential meaning for bridging the gap between immediate responses to new stressful conditions and the longer timescales of genetic adaptations.

11.10 – 12.25 Contributed Session C20: EvoDevo of cranial neural crest and dentition

ROOM A

Chairs: Moya Smith and Georgy Koentges

C20-01 An epithelial stem cell niche and a core gene network regulate continuous tooth regeneration in sharks

Martin, Kyle (University of Sheffield, GBR); Rasch, Liam (University of Sheffield, GBR); Fraser, Gareth (University of Sheffield, GBR)

Evolution has produced a wide diversity of dental phenotypes in vertebrates, each to suit particular trophic adaptations. Using the comparative method it is possible to uncover the developmental and genetic basis for this variation. While detailed studies of tooth development in models with highly derived dental phenotypes can be productive, we still know little about the evolutionary origin of teeth or the developmental and genetic basis of tooth formation in the first jawed vertebrates prior to the deep divergence of osteichthyans (bony vertebrates) and chondrichthyans (sharks, rays, and chimaerids). Here we present data on development and continuous replacement of teeth in the shark *Scyliorhinus canicula*. We show that tooth development in *S. canicula* relies on a core gene regulatory network conserved across extant gnathostomes, and that this network is redeployed in perpetuity during the life-long "conveyor-belt" system of tooth production. We suggest this mechanism of continuous de novo development is facilitated in part by a specialized cell niche at the interface of the oral epithelium and permanent dental lamina, which has stem-like properties. In situ hybridization shows several canonical stem-cell markers are expressed in this niche, and dual pulse-chase experiments with brdU and edU suggest that slow-cycling label retaining cells (LRCs) are concentrated in this superficial region. Using dil we observe putative transit-amplifying cells leaving the epithelial stem-niche and contributing to the dental lamina. By comparing mechanisms of tooth development at both the genetic and cellular level in *S. canicula* to bony vertebrates we will develop a better understanding of the ancestral mechanism of tooth development in vertebrates and how this has been changed to produce the diversity of dental phenotypes observed in nature.

C20-02 Morphogenetics of coordinated tooth and jaw development and evolution in mammals

Boughner, Julia (University of Saskatchewan, Saskatoon, CAN); Raj, Muhammad (University of Saskatchewan, Saskatoon, CAN); Uppal, Jasmene (University of Saskatchewan, Saskatoon, CAN)

While vertebrates living and extinct show an astounding variety of tooth and jaw phenotypes, how teeth and jaws have evolved to remain matched and functional regardless of changes in shape, size and, in the case of teeth, number, is poorly understood. Understanding the

processes orchestrating the coordinated development of teeth and jaws offers insight into how these tissues also evolved in a coordinated way. Using a mouse model system, our aim here was to start to characterize the developmental-genetic processes that control the developmental timing and spatial organization of forming teeth such that they maintain "fit" within the growing jawbone. Guided by the working hypothesis that teeth and jaws do not directly coordinate each other's morphogenesis but rather develop via autonomous gene regulatory networks, we contrasted gene expression and protein products between the mandibular arches of dentate (p63+/+, p63+/-) and toothless (p63-/-) mouse mutants aged embryonic days (E) 10-E14. We complemented the above microarray, Q-PCR and Western blot assays with micro-CT imaging and 3D geometric morphometric analyses of odontogenesis alongside upper and lower jaw skeletogenesis in similarly aged mice (E10-18). Our results suggest that normal lower jaw morphogenesis is independent of the lower dentition while upper jaw development may not be; and that the lower dentition develops via a tooth-specific gene regulatory network that is exclusive of the mandible. Thus far our ongoing study supports a high level of developmental-genetic autonomy between tooth and jaw tissues. While the specific developmental mechanisms that coordinate dental-gnathic morphogenesis remain to be characterized, our work implies that, ultimately, rigorous selection for functionally integrated teeth and jaws is the process driving the coordinated macroevolution of these oral tissues in mammals, and possibly in other vertebrate groups.

C20-03 Quantitative modeling of dental stem cell niche evolution and constant change in tooth height over 50 million years

Mushegyan, Vagan (University of California San Francisco, CA, USA); Eronen, Jussi (University of Helsinki, FIN & Senckenberg Research Institute and Natural History Museum, Frankfurt, GER); Lawing, Michelle (Texas A&M University, College Station, TX, USA); Sharir, Amnon (University of California San Francisco, CA, USA); Janis, Christine (Brown University, Providence, RI, USA); Klein V., Ophir D. (University of California San Francisco, CA, USA)

Novel traits allow species to adapt to changes within their ecological niches and to enter new adaptive zones. An example of such innovation is the evolution of adult dental stem cell niches in rodents, which results in continuously growing (hypselodont) molars. To determine the patterns of rodent molar stem cell evolution, we examined 3500 North American rodent fossils, ranging from 50 million years ago (mya) to 2 mya and conducted a molecular clock analysis of extant rodent species. In addition, we asked to what extent a simple Markov simulation model of constant increase in tooth height over time could explain the evolution of stem cells. The fossil record revealed that hypselodont phenotypes emerged through intermediate forms of increasing crown height, which served as convergent stepping-stones for the evolution of new stem cell niches. Our simple model required only two parameters to replicate the progressive increase in tooth height observed in the fossil record over 50 million years. Our data suggest that gradual and quantitative changes in crown height under a common evolutionary pressure led to emergence of the adult stem cell niche and predict that hypselodonty will eventually become the exclusive phenotype in rodents.

C20-04 Cellular cartilage predates vertebrates and was coopted by the neural crest during vertebrate head skeleton evolution

Jandzik, David (University of Colorado at Boulder, CO, USA); Garnett, Aaron T. (University of Colorado at Boulder, CO, USA); Square, Tyler A. (University of Colorado at Boulder, CO, USA); Cattell, Maria V. (University of Colorado at Boulder, CO, USA); Yu, Jr-Kai (Academia Sinica, Taipei, TWN); Medeiros, Daniel M. (University of Colorado at Boulder, CO, USA)

A defining feature of vertebrates is a pronounced head supported and protected by a cellularized endoskeleton. In the first vertebrates, it was likely built of collagenous cellular cartilage, which forms the embryonic skeleton of all vertebrates and the adult skeleton of modern jawless and cartilaginous fish. In the head, most cellular cartilage is derived from the neural crest. Because collagenous cellular cartilage and neural crest cells (NCC) have never been described in invertebrates, the appearance of NCC-derived cellular cartilage is considered a turning point in vertebrate evolution. We show that a skeletal tissue with the defining features of vertebrate cellular cartilage forms transiently in larvae of the invertebrate chordate, amphioxus. We also present evidence that a key regulator of collagenous cellular cartilage development, *SoxE*, gained new *cis*-regulatory sequences during vertebrate evolution to direct its novel expression in NCC. These results suggest that the origin of the vertebrate head skeleton was not dependent on the evolution of a new skeletal tissue, but by the spread of this tissue throughout the head. We further propose that the evolution of *cis*-regulatory elements near an ancient regulator of cartilage differentiation was a major factor in the origin and diversification of the vertebrate head skeleton.

C20-05 Afferent projections mirror the evolutionary origins of trigeminal sensory neurons

Butts, Thomas (King's College, London, GBR); Graham, Anthony (King's College, London, GBR)

Somatosensory sensation (of pain, touch, and position) in the face has proved crucial to the success of vertebrates and especially gnathostomes. It is mediated through the trigeminal sensory system. Developmentally, trigeminal neurons comprise three distinct cell populations:

those derived from placodes, from neural crest, and from the dorsal epithelium of the midbrain. The latter go onto form the mesencephalic trigeminal nucleus (MTN) that sits within the posterior midbrain. This complex development is unique amongst all ganglia, and makes the trigeminal an ideal model system for examining the evolution of sensory neurogenesis in vertebrates. In order to investigate the complexity within this system, we have fate-mapped the various distinct neuronal populations in chick and mouse and have shown that there is no meaningful relationship between developmental origin and afferent projection pattern for both placode- and crest-derived neurons. In contrast, by experimentally inducing a fate switch of trigeminal motor neurons from trigeminal to facial identity, we show that mesencephalic trigeminal afferent projections depend upon the trigeminal identity of motor neurons in the mandibular branch. Thus, the trigeminal sensory system reflects the complex evolutionary history of development in the head: anatomical specificity of afferent projection is confined to the MTN, whose projection into the lower jaw evolved after the split between agnathans and jawed vertebrates.

11.10 – 12.25

Contributed Session C21: Quantitative EvoDevo in model and non-model organisms I

ROOM B

Chair: Philipp Mitteroecker

C21-01 Adaptive integration in the human pelvis

Fischer, Barbara (Centre for Ecology and Evolutionary Synthesis, Oslo, NOR); Mitteroecker, Philipp (University of Vienna, AUT)

Compared to other primates, childbirth in humans is remarkably difficult. The evolution of upright walking, which required substantial change in pelvis shape, and the subsequent evolutionary increase of brain size led to the "obstetric dilemma": The head of the human fetus is large relative to the birth-relevant dimensions of the pelvis. It has been hypothesized that modern human pelvic morphology has therefore evolved as a compromise between being shaped for upright walking and giving birth to large-headed neonates. In this study we argue that the two essential functions of modern human pelvic morphology — bipedal movement and giving birth — are closely linked to stature and head circumference. Given the considerable heritability of both stature and head circumference, we hypothesize that pelvis shape has coevolved with stature and head circumference and that this correlated selection pressure on pelvis, head circumference, and stature has given rise to patterns of adaptive integration in these structures. We test this hypothesis by a geometric morphometric analysis using fine-resolution 3D landmark data from 99 human skeletons.

C21-02 Grasping flexibility: Evolutionary modularity and developmental origin of carapace integration in Chelonians

Djurakic, Marko (University of Novi Sad, SRB); Herrel, Anthony (UMR 7179 CNRS/MNHN, Paris, FRA); Jojic, Vida (Institute for Biological Research "Sinisa Stankovic", University of Belgrade, SRB)

Extant turtles, with minor exceptions, exhibit astonishing conservatism of bony shell parts in contrast to extreme variation in size and shape of the shell as a whole. As such, one might expect a certain degree of developmental flexibility among parts of the shell acting as semi-independent units or modules within functionally integrated shell. Utilizing 3D geometric morphometric techniques we quantified carapace shape of 168 adult turtles belonging to 47 species covering both Cryptodira and Pleurodira suborders. The first phylogenetic principal component (accounting for 42 % of the total shape variation) clearly showed a gradient from terrestrial, semi-aquatic to aquatic species suggesting that carapace shape is considerably constrained by environmental media and highlighting its adaptive role. Several modularity hypotheses, proposed based on fossil and developmental data, were tested using RV and multi-set RV coefficients (Klingenberg 2009). Taking into account phylogenetic structure of the data (analyses based on independent contrasts covariance matrix), we obtained statistical support for a two- to five-module organization of the turtle carapace. However, the best supported hypothesis partitions carapace into four semi-independent units that correspond to nuchal, costal, vertebral and marginal+pygial+suprapygial regions of the carapace. This pattern of modularity held up for the asymmetry covariance matrix as well. Furthermore, the developmental origin of carapace integration was assessed by an explicit comparison between the covariance matrices of fluctuating asymmetry and independent contrasts. This analysis yielded a moderate but highly significant matrix correlation suggesting that direct developmental interactions among carapace traits play a profound role in determining its shape variation as a whole. From these results we draw the following main conclusions. First, particular parts of the turtle's carapace act as semi-independent units likely enabling shell shape adaptations to differential environmental demands. Second, carapace integration is developmentally controlled by direct interactions, possibly inductive signaling. Finally, the observed pattern of modularity and integration, as well as the development of the carapace, is likely preserved in turtles before the split between Cryptodira and Pleurodira (220-157 MYA). Moreover, our results are in agreement with the folding theory of shell formation (Kuratani et al. 2011) and its extended version (Lyson et al. 2013) according to which developmental sequences that form the carapace during ontogeny resemble transitional events on a macroevolutionary scale.

C21-03 Systematic knock-down analysis of the gap gene network in the scuttle fly *Megaselia abdita* reveals quantitative system drift

Wotton, Karl (Center for Genomic Regulation, Barcelona, ESP); Jimenez-Guri, Eva (Center for Genomic Regulation, Barcelona, ESP); Crombach, Anton (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

Understanding development is often restricted by our inability to account for the expression dynamics and interactions of all relevant regulatory factors. One of the few developmental systems to reach such a comprehensive level of analysis is the gap gene system of *Drosophila melanogaster*, where saturation mutagenesis has identified all relevant factors, while genetic/molecular studies and mathematical modelling have subsequently refined our understanding of the dynamics of the system. Recreating such studies in non-model organisms represents a significant challenge to evo-devo. To address this problem we have carried out a systematic analysis of the gap gene system in the scuttle fly, *Megaselia abdita* (Phoridae). We first created detailed staging schemes and data processing pipelines to quantify the wild-type dynamics of gap gene expression at high spatial and temporal resolution. We then used RNA interference to systematically assess the sensitivity of the pattern forming capability of the network to gene knockdown. We find that expression dynamics differ significantly between *Drosophila* and *Megaselia* but converge to equivalent patterns just before gastrulation. In the latter, anterior stable gap gene boundaries now shift, while in the posterior, gap boundaries shift to the anterior with a marked time delay, which is compensated for by stronger shifts. Our knockdown analysis reveals a number of regulatory changes affecting the strength of gap gene cross-repression that contribute to these altered dynamics. Such quantitative changes in regulatory mechanisms have not yet been documented in mechanistic detail for any other developmental process. They are likely to be a common feature of developmental system drift.

C21-04 Evolution of skull shape in Triturus newts: An ontogenetic and phylogenetic perspective

Ivanovic, Ana (University of Belgrade, SRB); Cvijanovic, Milena (Institute for Biological Research "Sinisa Stankovic", Belgrade, SRB); Arntzen, Jan W. (Naturalis Biodiversity Center, Leiden, NDL); Zelditch, Miriam (University of Michigan, MI, USA)

Comparative studies of closely related species provide insights into the mechanisms responsible for morphological diversification. We explored ontogenetic and phylogenetic changes in skull shape using the monophyletic group of eight species that make up the salamander genus *Triturus*. Their well-studied phylogenetic relationships and the marked differences in ecological preferences make this genus a model system for the study of morphological evolution. Moreover, their biphasic life history with aquatic larvae and terrestrial adults, including an abrupt morphological change at metamorphosis, gives the opportunity to explore the effect complex life-cycles may have on the evolution of ontogeny and the generation of diversity. We found that the similarities among skull shapes reflect recency of common ancestry. When phylogenetic independent contrasts were used, skull size and shape were significantly correlated, indicating evolutionary allometry. However, analyses of the ontogenetic trajectories of post-metamorphic changes in skull size and shape show that complex changes — not simple allometric scaling — produced the morphological diversification in studied species.

C21-05 Evolution of salamander limbs: Influence of different functional demands of larvae and adults

Vukov, Tanja (University of Belgrade, SRB); Üzüm, Nazan (Adnan Menderes Universitesi, Aydin, TUR); Urosevic, Aleksandar (University of Belgrade, SRB); Slijepcevic, Maja (University of Belgrade, SRB); Tomasevic Kolarov, Natasa (University of Belgrade, SRB)

Salamanders have complex life cycles with a variety of life histories and developmental modes. In general, their biphasic cycle contains an aquatic larval form with limbs that are developing while in use and a metamorphosed terrestrial adult form with limbs that are functional in aquatic and terrestrial environments. The transition between two phases, metamorphosis, separates two main ontogenetic stages. Although limbs do not undergo drastic changes through process of metamorphosis, different constraints for locomotion in two main ontogenetic stages can influence adult limb morphology. In other words, adult limb morphology can be driven by functional demands in early life or/and by the selection for adult limb functional traits. We examine the evolution of limb skeletal morphologies in 18 salamander species in light of the relative impact of history (phylogeny) and different developmental modes and functional demands of the larvae and adults. We use phylogenetic comparative methods to relate limb skeletal morphometrics to developmental modes and life history features of larvae and adults (presence of larvae, direct development, stream-type larvae, pond-type larvae, bottom dwellers larvae, pelagic larvae, aquatic phenology of adults-length of aquatic phase). Results show influence of phylogeny and higher influence of functional demands of adults comparing to larvae on morphological evolution of limbs in salamanders.

11.10 – 12.25 **Contributed Session C22:**
Less is more: Loss of gene functions as a driving force of developmental evolution

ROOM C1

Chairs: Cristian Cañestro and Ingo Braasch

C22-01 The repertories of developmental transcription factors in sponges were shaped by extensive independent gene loss events

Fortunato, Sofia (University of Bergen, NOR); Adamski, Marcin (University of Bergen, NOR); Adamska, Maja (University of, NOR)

The genome of the demosponge *Amphimedon queenslandica* revealed a limited repertoire of developmental transcription factors (TFs) in comparison to cnidarians and bilaterians. This finding supported the view that increasing morphological complexity in animal lineages correlated with increasing developmental toolkit complexity. We have sequenced genomes of several representatives of calcareous sponges, a poriferan lineage that has separated from demosponges around the time demosponges separated from the lineage leading to cnidarians and bilaterians. In this study, by performing phylogenetic analyses of a wide range of TF gene families, we found that the gene complement in calcisponges dramatically differs from the one in *Amphimedon*. Our study shows that sponge genomes have undergone substantial gene loss of TF gene subfamilies. This suggests that many of the developmental TF gene families were established early in evolution, rather than in the last common ancestor of cnidarians and bilaterians, as previously assumed. In addition, many TFs found in ctenophores are not present in any of the analysed sponges and vice versa. This suggests severe gene loss in either of the two phyla, depending on their branching order. Our results support the hypothesis that the last common ancestor of extant animals was complex, and it was equipped with a more diverse developmental toolkit of TFs than extant sponges.

C22-02 Lens defects in *Astyanax mexicanus* blind cavefish: Focus on crystallins evolution and function

Hinaux, Hélène (CNRS Gif-sur-Yvette, FRA); Blin, Maryline (CNRS Gif-sur-Yvette, FRA); Fumey, Julien (CNRS Gif-sur-Yvette, FRA); Legendre, Laurent (CNRS Gif-sur-Yvette, FRA); Casane, Didier (CNRS Gif-sur-Yvette, FRA); Rétaux, Sylvie (CNRS Gif-sur-Yvette, FRA)

The fish *Astyanax mexicanus* presents, within the same species, several populations of river-dwelling surface fish and blind cave-living fish. In blind cavefish, the eyes first develop almost normally during embryogenesis. But 40 hours after fertilization (hpf), after the embryo has hatched, the lens enters apoptosis, which triggers the progressive degeneration of the entire eye. The mechanism leading to lens apoptosis is unknown. Before apoptosis, the lens expresses early differentiation markers correctly. We thus searched for possible late differentiation defects that would be causal in cavefish lens differentiation. We surveyed the cavefish and surface fish transcriptomes to uncover Astyanax crystallins, which are major lens structural components. These proteins are less polymorphic and accumulate more mutations in cavefish than in surface fish, suggesting relaxed selection at these loci in cavefish. Besides, at least 5 crystallins are not expressed correctly in cavefish, based on in situ hybridization and qPCR data. The functional role of two crystallin candidates was therefore tested by morpholino/transgenesis approach. As experimental manipulations of single crystallins do not impact lens development, we propose that a combinatorial effect of all the crystallins defects could trigger lens apoptosis. Thus, loss of crystallin expression/function would be part of the developmental mechanisms by which cavefish lost their eyes.

C22-03 Evolution and development of the bifurcated axial skeletal system in the twin-tail goldfish

Abe, Gembu (Academia Sinica, Yilan, TWN); Lee, Shu-Hua (Academia Sinica, Yilan, TWN); Chang, Mariann (Academia Sinica, Yilan, TWN); Liu, Shih-Chieh (Academia Sinica, Yilan, TWN); **Ota, Kinya** (Academia Sinica, Yilan, TWN)

Twin-tail goldfish possess a laterally bifurcated caudal axial skeleton. The scarcity of such highly modified caudal axial skeletal systems in nature suggests that a rare mutation, which drastically altered the developmental mechanisms underlying axial skeleton formation, may have occurred during goldfish domestication. However, little is known about the molecular development of the bifurcated caudal skeleton in twin-tail goldfish. Here, we report that the bifurcated caudal skeletal system arises from a mutation in the chordin gene, which affects embryonic dorsal-ventral patterning. Morphological observation, backcross analysis and functional assays were used to demonstrate that formation of the laterally bifurcated caudal axial skeleton requires a stop-codon mutation in one of two recently duplicated chordin genes. Analysis of gene expression patterns suggests that the ventral tissues of the twin-tail goldfish are increased in size, and these tissues form the embryonic bifurcated fin fold. Moreover, unlike previously described *chordin*-deficient vertebrate embryos, anterior-dorsal neural tissues were not reduced in twin-tail goldfish embryos; this presumably prevented by the presence of a duplicated chordin gene. Furthermore, analysis of Chinese archives suggests that the developmental changes might have occurred within an approximately 600-year period of domestication. Based on our recent findings, we discuss how the gene duplications and accumulated mutations have modified the axial skeletal system in the vertebrate lineage.

C22-04 A mollusk retinoic acid receptor (RAR) ortholog sheds light on the evolution of ligand binding

Schubert, Michael (Laboratoire de Biologie du Développement de Villefranche-sur-Mer, FRA)

Nuclear receptors (NRs) are transcription factors that regulate networks of target genes in response to small molecules. There is a strong bias in our knowledge of these receptors, since they were mainly characterized in classical model organisms, mostly vertebrates. Therefore, the evolutionary origins of specific ligand-receptor couples still remain elusive. Here, we present the identification and characterization of a retinoic acid receptor (RAR) from the mollusk *Nucella lapillus* (NlRAR). We show that this receptor specifically binds to DNA response elements organized in direct repeats as a heterodimer with the retinoid X receptor (RXR). Surprisingly, we also find that NlRAR does not bind all-trans retinoic acid (RA) or any other retinoid we tested. Furthermore, NlRAR is unable to activate the transcription of reporter genes in response to stimulation by retinoids and to recruit co-activators in the presence of these compounds. 3D modeling of the ligand-binding domain of NlRAR reveals an overall structure that is similar to vertebrate RARs. However, in the ligand-binding pocket (LBP) of the mollusk receptor, the alteration of several residues interacting with the ligand has apparently led to an overall decrease in the strength of the interaction with the ligand. Accordingly, mutations of NlRAR at key positions within the LBP generate receptors that are responsive to retinoids. Altogether, our data suggest that, in mollusks, RAR has lost its affinity for all-trans RA, highlighting the evolutionary plasticity of its LBP. When put in an evolutionary context, our results reveal new structural and functional features of NRs validated by millions of years of evolution that were impossible to reveal in model organisms.

C22-05 Darwin's "living fossil" as a new model: Spotted gar and the genomic basis of vertebrate EvoDevo

Braasch, Ingo (University of Oregon, Eugene, OR, USA); Batzel, Peter (University of Oregon, Eugene, OR, USA); Loker, Ryan (University of Oregon, Eugene, OR, USA); Amores, Angel (University of Oregon, Eugene, OR, USA); Fontenot, Quenton (Nicholls State University, Thibodaux, LA, USA); Ferrara, Allyse (Nicholls State University, Thibodaux, LA, USA); Postlethwait, John H. (University of Oregon, Eugene, OR, USA)

Spotted gar (*Lepisosteus oculatus*) — a holostean rayfin fish that diverged from teleost fish shortly before the teleost genome duplication (TGD) and one of Darwin's defining examples of "living fossils" — holds clues to the ancestry of vertebrate gene functions and provides connectivity among vertebrate genomes: The TGD had major impact on the evolution of teleost genomes and gene functions. Furthermore, the earlier two vertebrate genome duplications (VGD1/2) complicate the analysis of vertebrate gene family history and gene function evolution. Following the genomic "big bang" of genome duplications, subsequent lineage-specific genome reshuffling and loss of gene duplicates can obscure the distinction of orthologs and paralogs across lineages, leading to false conclusions about the origin of vertebrate genes and their functions. Using a chromosome-level genome assembly of spotted gar, we show that gar has retained many paralogs from VGD1/2 that were differentially lost in teleosts and lobefins (coelacanth, tetrapods). In addition, spotted gar can be reared as a laboratory Evo-Devo model enabling the functional testing of hypotheses about the origin of rayfin and lobefin gene activities without the confounding effects of the TGD. The spotted gar genome sequence also facilitates the identification of *cis*-regulatory elements shared between teleosts and tetrapods, revealing hidden orthology among regulatory elements that cannot be established by direct teleost-tetrapod comparisons. Using whole genome alignments of teleosts, spotted gar, coelacanth, and tetrapods, we identify conserved non-coding elements (CNEs) that were gained and lost at different times during vertebrate evolution. This information enables us to study on a genome-wide scale the role of regulatory sub- and neofunctionalization after the TGD and helps to infer targets of *cis*-regulatory elements that we test *in vivo* using transgenic reporter assays. In conclusion, this "living fossil" links teleost genomes to tetrapod biology.

11.10 – 12.25 Contributed Session C23: EcoEvoDevo: Symbiosis and epigenetic inheritance

ROOM C2

Chairs: Scott Gilbert and Yoav Soen

C23-01 Development of symbiotic organ: Hox genes regulate development of bacteriome and localization of bacterial symbiont in seed bug *Nysius plebeius*

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Symbiotic microbes often specifically infect and localize in host tissues in accordance with development of their hosts. Hemipteran insects, such as aphids and leafhoppers, are known to harbor nutrient-supplying symbionts in symbiotic cells, called "bacteriocytes", and transmit the symbionts to the next generation through ovary. During embryogenesis, a cell population accepts the symbiotic bacterium, differentiates into the bacteriocytes and finally develops into a large organ, called "bacteriome". While numerous histological descriptions have been

documented so far (Buchner 1965), no study has addressed genetic and molecular mechanisms underlying the development of bacteriome. Many phytophagus stinkbugs (Hemiptera; Heteroptera; Pentatomomorpha) harbor beneficial symbiotic bacteria in crypts of their posterior midgut and orally acquire them every generation. Meanwhile, a few genera of the family Lygaeidae (seed bugs) lack midgut crypts, but acquired novel bacteriomes harboring intracellular symbionts. We have previously identified an endosymbiotic association between *Nysius* seed bugs and bacterial symbionts "*Candidatus Schneideria nysicola*" in a pair of bacteriomes. Genomic analyses and antibiotic treatment indicated an obligate nature of the *Nysius-Schneideria* symbiosis. In an attempt to understand the developmental basis of *Nysius* bacteriome, we described unique organogenesis of bacteriomes in embryos of *N. plebeius* by whole-mount FISH. Schneideria cells infected presumptive bacteriocytes in abdominal segments before katatrepsis. We then conducted parental RNAi of several candidate genes, thereby identifying a single *Hox* gene that is crucial for bacteriome development. By RNAi of this gene, the bacteriomes were lost and *Schneideria* failed to infect the germband. Expression of this gene was detected in the bacteriocytes even before symbiont infection. Suppressing other *Hox* genes altered the number and localization of bacteriomes. Based on these results, we discuss the molecular mechanism and evolution of the bacteriome in *N. plebeius*.

C23-02 Ancestral developmental potential facilitates parallel evolution of a novel supersoldier caste in the hyperdiverse ant genus Pheidole

Rajakumar, Rajendhran (McGill University, Montreal, QC, CAN); San Mauro, Diego (McGill University, Montreal, QC, CAN); Dijkstra, Michiel B. (McGill University, Montreal, QC, CAN); Huang, Ming H. (University of Arizona, Tucson, AZ, USA); Wheeler, Diana E. (University of Arizona, Tucson, AZ, USA); Hiou-Tim, François (McGill University, Montreal, QC, CAN); Khila, Abderrahman (McGill University, Montreal, QC, CAN); Cournoyea, Michael (McGill University, Montreal, QC, CAN); Abouheif, Ehab (McGill University, Montreal, QC, CAN)

Complex worker caste systems have contributed to the evolutionary success of advanced ant societies; however, little is known about the developmental processes underlying their origin and evolution. We combined hormonal manipulation, gene expression, and phylogenetic analyses with field observations to understand how novel worker subcastes evolve. We uncovered an ancestral developmental potential to produce a "supersoldier" subcaste that has been actualized at least two times independently in the hyperdiverse ant genus *Pheidole*. This potential has been retained and can be environmentally induced throughout the genus. Therefore, the retention and induction of this potential have facilitated the parallel evolution of supersoldiers through a process known as genetic accommodation. Recurrent phenotypes reflecting ancestral potentials have long been recognized as widespread in plants and animals. Because of the lack of empirical evidence, however, the evolutionary significance of these recurrent phenotypes has been underappreciated. We show that the recurrent induction of ancestral developmental potential is a source of adaptive variation for selection that may facilitate the adaptive and parallel evolution of phenotypes. Environmental induction of ancestral developmental potentials must therefore be included in any extended synthesis of evolutionary theory.

C23-03 Investigating the role of genetic assimilation in an adaptive radiation

Gunter, Helen (University of Konstanz, GER); Karner, Immanuel (University of Graz, AUT); Schneider, Ralf (University of Konstanz, GER); Sturmbauer, Christian (University of Graz, AUT); Meyer, Axel (University of Konstanz, GER)

Explosive, concurrent speciation events within a lineage that typically result in derived, ecologically diverse species compared to the ancestral state, are termed adaptive radiations, and constitute ideal models to investigate major evolutionary mechanisms. It was proposed that adaptive radiations may occur when ancestral lineages display high levels of developmental plasticity in adaptive traits, facilitating ecologically-driven phenotypic diversification, which can then become genetically fixed through the mechanism of genetic assimilation. In our research we investigate whether genetic assimilation played a role in the adaptive radiation of East African cichlid fishes, which belongs to the most specious family of vertebrates. Through using a split brood experimental design, we investigated whether a derived, ecologically specialized species from within the Lake Victorian radiation (*Haplochromis ishmaeli*), displays lower levels of diet-induced phenotypic plasticity than lineages that are basal to it and did not undergo adaptive radiation (*Astatoreochromis alluaudi* and *Astatotilapia burtoni*). Morphologically, both basal lineages displayed higher levels of plasticity than *H. ishmaeli*. Additionally, qRT-PCR analyses of previously identified "plasticity genes" indicated that this reduced level of plasticity is associated with a reduced reaction norm in candidate gene expression for *H. ishmaeli* when compared to *A. burtoni* and *A. alluaudi*. Our results suggest that genetic assimilation may have occurred during the Lake Victorian adaptive radiation, a hypothesis that can be further tested through examining the evolution of transcription factor binding sites of our candidate genes for species both within, and basal to this cichlid adaptive radiation.

C23-04 Genetic assimilations: How they impact blind variation

Danchin, Etienne (Evolution & Diversité Biologique, UMR 5174, CNRS, UPS, ENFA, Toulouse, FRA); **Pocheville, Arnaud** (University of Pittsburgh, PA, USA)

In this contribution, we wish to clarify several points regarding genetic assimilation in evolutionary biology. We mean here by genetic assimilation a situation where some phenotypic variations that are not initially caused by environmental events eventually get caused by genetic variations (cause meaning here a difference maker). The concept of genetic assimilation has given rise to some controversy (Pigliucci et al. 2006), having for instance been considered as an intended contradiction to the Modern Synthesis that can be explained as merely resulting from a selective process (de Jong 2005). We will first sketch a brief historical overview of the ideas similar to genetic assimilation (e.g., Baldwin 1896), and of its framing in genetic terms (e.g., Waddington 1942, Schmalhausen 1949). We will then discuss two recent models of genetic assimilation that make use of non-genetic inheritance mechanisms (Klironomos et al. 2013; Danchin et al. in prep.). We will show how genetic assimilation can occur at two levels: the population, or the individual. We will propose that genetic assimilation occuring at the level of the individual can be adaptive without necessarily resulting from a previous selective process, thus leading to evolvability as an exaptation of plasticity. This will lead us to discuss the role of blind variation and selection in explanations of adaptation, and the compatibility of (non)-genetic assimilation with the Modern Synthesis.

C23-05 The propensity interpretation of fitness and the Modern Synthesis

Chiu, Lynn (University of Columbia-Missouri, MO, USA)

Griffiths and Gray (2001) argue that the orthodox interpretation of fitness applies equally well to Developmental Systems Theory (DST). I argue that it does not. The problem lies not with DST, but with the orthodox view of fitness. I argue that the orthodox interpretation is based on a Modern Synthesis assumption: the processes that generate developmental varieties are independent of the conditions of selection (Lewontin 2000). This assumption is not true for many developmental systems. The orthodox position, formally called the Propensity Interpretation of Fitness (PIF), defines fitness as the probabilistic disposition of an evolutionary unit to propagate itself (Brandon 1978; Mills and Beatty 1979; Pence and Ramsey 2013). Since a developmental system — the package of developmental resources that reliably reconstruct a life cycle — is such an evolutionary unit, Griffiths and Gray state that, clearly, the PIF analyses carry over. However, PIF prescribes two ways to measure fitness. Just as differences in the solubility of salts is found by examining their physical differences or by observing differences after dissolving them in a solution, the differences in fitness-as-propensity can be found by examining the differences in biological and ecological properties or by measuring actual reproductive differences. The analogy between fitness and garden-variety dispositions such as solubility assumes that differences in the physical basis of fitness can be considered independently and separately of their developmental background. This is equivalent to the Modern Synthesis assumption that the processes generating developmental variation can be considered independently of their selective environment. Yet, Saltz and Nuzhdin (2013) show that genetic variation in habitat selection and the creation of social environments implies that different genotypes of the same population experience different developmental environments, which in turn could influence plastic phenotypes. Therefore, the selective environment — the total influence of developmental environments on fitness differences — cannot be considered independently of these variations and their generation. In other words, the complex interplay between developmental resources rejects statistical parsing into the main effects of "evolutionary unit" and "developmental background," preventing the direct application of PIF. PIF arose to answer two philosophical problems: is natural selection tautological? What is the connection between ecological adaptedness and the mathematical notion of fitness? However, its application is limited by a hidden Modern Synthesis assumption. Following the call for an updated "eco-evo-devo" definition of natural selection (Blute 2008; Walsh 2009), there is a need for an updated eco-evo-devo interpretation of fitness.

11.10 – 12.25 Contributed Session C24: Origin and diversification of regeneration

ROOM D

Chair: Jeremy Brockes

C24-01 Using sponges to investigate the evolution of stem cell gene regulatory networks

Revilla-i-Domingo, Roger (Max F. Perutz Laboratories / University of Vienna, AUT); Steudle, Friederike (Max F. Perutz Laboratories / University of Vienna, AUT); Raible, Florian (Max F. Perutz Laboratories / University of Vienna, AUT)

Across all phyla of multicellular animals, stem cells are fundamental to sustain tissue homeostasis and regeneration. The unique capabilities of stem cells must be controlled by a complex program encoded in the genome, in the form of a gene regulatory network. The question we want to address is: what were the gene regulatory network changes that gave rise to stem cells during the evolution of multicellular animals? To address this question we are studying how somatic stem cells are regulated in the sponge, *Suberites domuncula*. The unique evolutionary position of sponges, as one of the most basal metazoan phyletic lineages, makes them ideal organisms to unravel ancestral

metazoan regulatory mechanisms. In addition, sponges are long lived, and have impressive regenerative capabilities, making their somatic stem cells a great model for metazoan stem cell biology. Archeocytes have been suggested to be totipotent stem cells in sponges. In at least one sponge species, archeocytes have been shown to be proliferative, to express the stem cell marker piwi, and to be required for regeneration from dissociated cells. However, to proof that arecheocytes are totipotent stem cells will require to test the self-renewal and differentiation potential of individual archeocytes. Our fist goal is to test the hypothesis that indeed archeocytes are totipotent stem cells, responsible for adult sponge homeostasis and regeneration. We have established a regeneration assay, where small explants of adult sponge tissue are able to attach and re-grow on glass slides. Our experiments indicate that this process depends on cell proliferation: Immunostaining with anti-Phospho-Histone H3 antibody, shows increased proliferation during regeneration, and gamma irradiation is able to prevent the regeneration process. We are now working towards: (1) Showing that irradiated explants lack archeocytes; (2) using this assay to test stem cell functions, by transplanting labeled candidate stem cells into the irradiated explants. In the long term we expect that our regeneration assay will serve as the basis for establishing functional genomics techniques, and for unraveling the gene regulatory network that controls stem cell functions in sponges.

C24-02 Transcriptional activation of Tgf-beta and Wnt pathways during whole body regeneration in sponges

Adamski, Marcin (University of Bergen, NOR); Laplante, Mary (University of Bergen, NOR); Liu, Jing (University of Bergen, NOR); Bråte, Jon (University of Oslo, NOR); Leininger, Sven (University of Bergen, NOR); Leon Florian, Luis Anthony (University of Bergen, NOR); Ereskovsky, Alexander (Mediterranean Institute of Marine and Terrestrial Biodiversity and Ecology (IMBE), Marseille, FRA); **Adamska, Maja** (University of Bergen, NOR)

Regenerative capacity of sponges, allowing them to recover complete bodies from fragments of tissue or even dissociated cells, is truly remarkable, and not many animals can match this potential. While continuously growing amount of information is available on genes involved in regeneration of model species, including cnidarians, acoels, planarians and chordates, nothing is known about molecular aspects of sponge regeneration. We are studying regeneration of *Sycon ciliatum*, a calcareous sponge, which has capacity to restore complete bodies from narrow rings of tissue. We applied RNA-Seq technology to identify genes involved in all steps of the regeneration process, beginning with wound healing and leading to complete recovery of the lost structures. Restoration of the apical osculum equipped with contractile sphincter is complete within approximately 5 days. Within three hours after the cutting, over one thousand genes are differentially regulated. While overwhelming majority of these genes encode for novel proteins, components of two key metazoan developmental pathways, Wnt and TGF-beta, are among the significantly and dramatically upregulated transcripts. In particular, eight (out of 22 present in *Sycon*) TGF-beta ligands and six (out of 21) Wnt ligands, as well as effectors (Smads, Tcf) and modulators (SFRPs) of these pathways are differentially expressed either throughout the regeneration process or concomitantly with specific morphogenetic events. As in situ hybridization protocols developed for *Sycon* allow cell-specific resolution, we are now investigating precise expression patterns of the differentially expressed genes in order to gain insight in the cellular processes (especially migration and transdifferentiation) in which they might be involved. We already know that in intact sponges many, but not all, of these genes are specifically expressed or enriched in the apical (oscular) region, which was recently suggested to be homologous to the head organizer of cnidarians. Thus, in both sponges and eumetazoans, "organizer genes" are also involved in regeneration, suggesting a common genetic basis of regeneration processes across the animal kingdom.

C24-03 Unexpected capacity for the primary body plan regeneration in *Nematostella vectensis* embryos dissociated into single cells

Kozyreva, Anastasia (Lomonosov Moscow State University, Faculty of Biology, Moscow, RUS); Genikhovich, Grigory (University of Vienna, AUT); Technau, Ulrich (University of Vienna, AUT)

Most basally branching metazoans, have a remarkable capacity to regenerate missing body parts even after severe damage. Comparative analysis of regeneration with normal development may reveal the ancestral mechanisms of this phenomenon. The normal body plan is first established during embryogenesis. To test the regenerative capacity of embryonic cells we dissociated early gastrulae of *Nematostella vectensis* into single cells using Ca2+/Mg2+-free seawater. Upon reaggregation, aggregates were able to restore the pattern and to regenerate polyps with one or several mouth openings. To investigate the sequence of events leading to de novo axis formation, we studied the expression of several marker genes during 1-10 days after reaggregation. We found that in 30 minutes after the reaggregation cells expressing the oral markers Wnt1, Wnt4, Bra, FoxA, the aboral marker Fgfa1 and mid-body marker Wnt2 were randomly distributed and there were no signs of axial patterning at the molecular level. In 3 days molecular polarity was reestablished from randomized conditions and expression patterns were similar to those in normal development, but often multiplied. Double in situ hybridization showed that in an aggregate there were 2-3 times more areas of oral marker expressi-

on than of aboral marker expression. In line with this, we found that an aggregate forms usually more hypostomes than feet. Blastomere cutting experiments have previously revealed that only animal halves are able to restore the full pattern, suggesting that it contains organizing activity (Fritzenwanker et al. 2007). We therefore wished to test the regenerative capacity of animal and vegetative cells in embryonic aggregates. We found that aggregates derived from animal/oral cells of mid gastrulae were able to form primary polyps, but aggregates derived from the aboral cells formed only ciliated balls. However, when aboral cells obtained from eF1α::memOrange transgenic embryos were aggregated along with the wild-type oral cells, aboral cells took part in the formation of the oral structures. This indicates that aboral cells are able to change their prospective fate in the presence of oral cells. Then we tried to obtain aggregates composed of only prospective endo- or prospective ectodermal cells. Endodermal cells were almost unable to aggregate and could not regenerate polyp-like structures. By contrast, aggregates composed of only ectodermal cells formed primary polyps. This indicates that ectodermal cells could compensate for the loss of prospective endodermal cells, while endodermal cell were unable to differentiate ectodermal cells. In summary we have revealed that embryonic cells of *N. vectensis* are much more plastic than previously thought. Reaggregated cells of mid gastrulae are able to self-organize on molecular, cellular and tissue levels leading to the regeneration of a primary polyp.

C24-04 Posterior elongation in the annelid *Platynereis dumerilii* involves stem cells molecularly related to primordial germ cells

Gazave, Eve (CNRS — Institut Jacques Monod, Paris, FRA); Béhague, Julien (CNRS - Institut Jacques Monod, Paris, FRA); Laplane, Lucie (CNRS — Institut Jacques Monod, Paris, FRA); Demilly, Adrien (CNRS — Institut Jacques Monod, Paris, FRA); Balavoine, Guillaume (CNRS — Institut Jacques Monod, Paris, FRA); Vervoort, Michel (CNRS — Institut Jacques Monod, Paris, FRA)

Like most bilaterian animals, the annelid Platynereis dumerilii generates the majority of its body axis in an anterior to posterior temporal progression with new segments added sequentially. This process relies on a posterior subterminal proliferative body region, known as the "segment addition zone" (SAZ). We explored some of the molecular and cellular aspects of posterior elongation in *Platynereis*, in particular to test the hypothesis that the SAZ contains a specific set of stem cells dedicated to posterior elongation. We cloned and characterized the developmental expression patterns of orthologs of 17 genes known to be involved in the formation, behavior, or maintenance of stem cells in other metazoan models. These genes encode RNA-binding proteins (e.g., *tudor, musashi, pumilio*) or transcription factors (e.g., *myc, id, runx*) widely conserved in eumetazoans. Most of these genes are expressed both in the migrating primordial germ cells and in overlapping ring-like patterns in the SAZ, similar to some previously analyzed genes (*piwi, vasa*). The SAZ patterns are coincident with the expression of proliferation markers *cyclin B* and *PCNA*. EdU pulse and chase experiments suggest that new segments are produced through many rounds of divisions from small populations of teloblast-like posterior stem cells. The shared molecular signature between primordial germ cells and posterior stem cells in *Platynereis* thus corresponds to an ancestral "stemness" program.

C24-05 Early evolution of limb regeneration in tetrapods – evidence from a Palaeozoic amphibian

Fröbisch, Nadia (Museum für Naturkunde Berlin, GER); Bickelmann, Constanze (Museum für Naturkunde, GER); Witzmann, Florian (Museum für Naturkunde, GER)

Salamanders show the by far highest capacity of regeneration among tetrapods, including full limb regeneration throughout their entire lives. The latter has classically been regarded as derived for salamanders while most research effort has focused on its molecular mechanisms. Based on a pattern of abnormalities distinctive for irregular regeneration in extant salamanders, we show that a distant fossil relative of modern amphibians, the Lower Permian (300 Ma) temnospondyl *Micromelerpeton* was capable of fully regenerating its limbs. Variant patterns occur in preaxial, central, and postaxial positions of fore- and hind limbs and include various degrees of fusion in the digits along the proximo-distal axis resulting in enlarged metapodial elements and distal bifurcations, addition of adventitious digits, and reduction or increase of phalangeal numbers in the regenerated digits. The pattern of abnormalities in *Micromelerpton* are directly comparable to the variant morphological patterns in the limbs of extant salamanders, which have been demonstrated to be caused by limb regeneration, but do not occur as variants of normal limb development. The finding for the first time enables a deep time perspective of the evolution of limb regeneration in vertebrates based on first hand data from the fossil record. It not only demonstrates that regeneration is an ancient capacity among amphibians, but the likeness of abnormalities in *Micromelerpeton* and salamanders indicates that comparable molecular mechanisms for the organization of the regenerate in modern salamanders were already in place 300 Ma ago. Moreover, the phylogenetic distribution of regenerative capacity in paired appendages suggests that this may indeed be an ancient feature of bony fish (including tetrapods) and that the molecular mechanisms of regeneration may still be present in frogs and amniotes, albeit not usually used.

14.00 – 15.40 Symposium S22: 'NEPTUNE' ITN: The evolution of sensory systems in the marine environment

ROOM A

Organizers: Andrew Hejnol and Maria Ina Arnone
Chairs: Andrew Hejnol and Maria Ina Arnone

S22-01 Novel photoreceptors in brachiopod embryos

Passamaneck, Yale (University of Hawaii, Honolulu, HI, USA); Martindale, Mark (University of Florida, St. Augustine, FL, USA)

Photoreceptor cells in bilaterian animals are generally grouped into two classes based upon photoreceptive membrane morphologies — ciliary and rhabdomeric. This morphological dichotomy is further supported by the expression of different classes of opsins, as well other members of the signal transduction cascade, in ciliary and rhabdomeric photoreceptor cells. Based upon this dichotomy in morphology and expression, it is hypothesized that the two classes of photoreceptors evolved prior to the diversification of the Bilateria, and that the distinction between these two cell types has been maintained through evolution. Although both photoreceptor cell types are present in many bilaterian lineages, there appear to be clade-specific differences in their utilization, with protostomes possessing cerebral eyes with rhabdomeric photoreceptors, and deuterostomes having cerebral eyes with ciliary photoreceptors. We have found that unlike other protostomes, the cerebral eyes of the brachiopod *Terebratalia transversa* are composed of cells that have a ciliary morphology instead of a rhabdomeric morphology. Transcriptome based analysis reveals, however, that these ciliary photoreceptor cells express both ciliary and rhabdomeric opsins. In addition, we have found that the ciliary opsin as well as two Go class opsins are expressed during gastrulation stages at the apical plate, along with other components of the phototransduction cascade. Our results evidence that at least two distinct photoreceptor systems are deployed during the embryonic and larval development of Terebratalia, and suggest that the distinction between ciliary and rhabdomeric photoreceptor cells is more labile than previously hypothesized.

S22-02 Mechanism of phototaxis in *Platynereis* larvae and the origin of visual eyes

Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

Phototaxis is widespread among planktonic organisms, and can be found in the larval stages of sponges, cnidarians, protostomes and deuterostomes. The ability of zooplankton to find their preferred water depth depends on varying daily light conditions and developmental stage. Planktonic larvae often undergo a behavioral change, switching from positive phototaxis, characteristic of the post-hatching stages, to negative phototaxis, characteristic of later larval stages before settlement and metamorphosis. The marine annelid *Platynereis dumerilii* is an excellent laboratory model to study the mechanisms of larval phototaxis. *Platynereis* has a bentho-pelagic-life cycle with a pelagic larva that shows early positive and late negative phototaxis. The neuronal circuit and mechanism of early larval phototaxis is well understood: the larval eyespots, consisting of a shading pigment cell and a rhabdomeric photoreceptor cell, mediate this response. The eyespot photoreceptor directly innervates the ciliary band (prototroch). The mechanism and neural circuitry underlying negative phototaxis is unknown. To study the mechanism of negative phototaxis in *Platynereis* larvae we combined behavioral experiments, laser ablation, and transmission electron microscopy. Late *Platynereis* larvae have six eyes, the two eyespots and four additional dorsal eyes, precursors of the adult eyes. We characterized the role of these eyes in larval phototaxis, using laser ablations. Our electron microscopic reconstructions revealed how the eyes regulate motor output during phototactic turning. The *Platynereis* eye circuit shows the hallmarks of a simple visual system, including spatial light detection and contrast modulation, illustrating how image-forming eyes may have evolved via intermediate stages capable to contrast only a light and a dark pixel during phototaxis.

S22-03 Eye evolution: Common use and independent recruitment of genetic components

Vopalensky, Pavel (Academy of Sciences of the Czech Republic, Prague, CZE); Pergner, Jiri (Academy of Sciences of the Czech Republic, Prague, CZE); Kozmikova, Iryna (Academy of Sciences of the Czech Republic, Prague, CZE); **Kozmik, Zbynek** (Academy of Sciences of the Czech Republic, Prague, CZE)

Animal eyes can vary in complexity ranging from a single photoreceptor cell shaded by a pigment cell to elaborate arrays of these basic units, which allow image formation in compound eyes of insects or camera-type eyes of vertebrates. The evolution of the eye requires involvement of several distinct components-photoreceptors, screening pigment and genes orchestrating their proper temporal and spatial organization. Analysis of particular genetic and biochemical components shows that many evolutionary processes have participated in eye evolution. Multiple examples of co-option of crystallins, Galpha protein subunits and screening pigments contrast with the conserved role of opsins and a set of transcription factors governing eye development in distantly related animal phyla. The direct regulation of essential photoreceptor genes by these factors suggests that this regulatory relationship might have been already established in the ancestral photoreceptor cell. Common use and independent recruitment of genetic components will be discussed in the context of selected marine species.

S22-04 Opsins and the evolution of eyespots in the Acoelomorpha

Pang, Kevin (University of Bergen, NOR); Hejnol, Andreas (University of Bergen, NOR)

Nearly all animals are capable of photoreception. While the diversity of photoreception systems can vary from a single individual cell to a complex lens-including eye, the main light-sensitive molecule utilized is opsin, a G protein-coupled receptor. Using newly available transcriptomic and genomic data from previously unsampled animal groups, including acoels, nemertodermatids, gastrotrichs, nemerteans, and priapulids, we gain a better understanding of the evolution of the opsin gene family and its distribution across the Metazoa. In particular, we focus on the Acoelomorpha (Acoela + Nemertodermatida), a group of small, morphologically simple, worm-like animals, whose phylogenetic position remains uncertain. Within this group, some species have simple eyespots, while others are lacking. Using two easy to culture acoel species, *Convolutriloba macropyga*, which has juvenile and adult eyespots, and *Isodiametra pulchra*, which lacks eyespots, we examine opsin diversity and expression. Transcriptomic searches identify 6-7 opsin genes in each acoel species. Surprisingly, these acoel opsins form a clade that is sister-to or within the rhabdomeric opsin group, suggesting that a lineage specific diversification occurred within the acoels, which is something that has been seen previously in cnidarians and arthropods. Only one *C. macropyga* opsin gene is expressed in both juvenile and adult eyespots. The others are expressed in putative ectodermal sensory cells in the juvenile, and possibly in the eyes of adults. In comparison, all *I. pulchra* opsins are expressed in the central nervous system, including parts of the brain, as well as the neurite bundles that run along the body axis. Our data, along with morphological data from other acoel species and outgroup comparisons, would suggest that acoels evolved eyespots independently, possibly in relation to habitat and life history. The expression of opsins in both acoel species would suggest a direct input to the nervous system, and may be related to changes in behavior or circadian rhythms. However it remains to be shown that these opsins are directly involved in light detection.

14.00 – 15.40 Symposium S23: Quantitative EvoDevo in model and non-model organisms II

ROOM B

Organizers: Benedikt Hallgrimsson, Chris Klingenberg, Philipp Mitteroecker, and Ruth Flatscher
Chair: Benedikt Hallgrimsson

S23-01 Yin and yang of EvoDevo: Evolutionary transitions and developmental experiments

Jernvall, Jukka (University of Helsinki, FIN)

Contributions from experimental developmental biology have had a limited role in capturing the gradual aspects of evolutionary change. This discrepancy is particularly evident in research on mammalian teeth. To reconstruct the Tree of Life of extinct animals, teeth are often the only evidence that palaeontologists have at their disposal. Consequently, many detailed features of teeth are used to determine relatedness of fossil species. Yet, experiments on teeth typically produce saltational changes on the phenotype. To link changes in development to evolutionary transitions, experiments and computational modeling was used to gradually change mouse tooth shape and produce transitions observable in the fossil record.

S23-02 Wnt signalling underlies craniofacial variability in Lake Malawi cichlids

Parsons, Kevin (Institute of Biodiversity, Animal Health, and Comparative Medicine, Glasgow, GBR); Taylor, Trent (University of Massachusetts Amherst, MA, USA); Powder, Kara (University of Massachusetts Amherst, MA, USA); Albertson, R. Craig (University of Massachusetts Amherst, MA, USA)

Progress towards understanding adaptive radiations at the mechanistic level is still limited with regard to the proximate molecular factors that both promote and constrain evolution. We focus on the craniofacial skeleton and show that expanded Wnt/β-catenin signalling early in ontogeny is associated with the evolution of phenotypic novelty and ecological opportunity in Lake Malawi cichlids. We demonstrate that the mode of action of this molecular change is to effectively lock into place an early larval phenotype, likely through accelerated rates of bone deposition. However, we demonstrate further that this change toward phenotypic novelty may in turn constrain evolutionary potential through a corresponding reduction in craniofacial plasticity at later stages of ontogeny. In all, our data implicate the Wnt pathway as an important mediator of craniofacial form and offer new insights into how developmental systems can evolve to both promote and constrain evolutionary change.

S23-03 Using false flowers to study evolution of modularity and integration

Armbruster, W. Scott (University of Portsmouth, GBR)

Flowers are a key innovation in the evolution of higher plants, spurring diversity and success, yet the origin and refinement of the flower are shrouded in the fogs of a distant past, >150 my ago. Modularity and/or phenotypic integration of floral parts were probably early innovations and responsible, in part, for the efficient function of flowers and success of flowering plants. We can learn about the origins of floral modularity and integration by studying a more recent "reinvention" of

the perfect blossom (the pseudanthium or false flower, a flower-like inflorescence,) where the serial homologies of functional components are more readily apparent. To this end, we discuss aspects of integration and modularity of blossom and vegetative traits of *Dalechampia scandens* subjected to diverse environmental treatments. We find that blossom components that function together are phenotypically integrated, despite being structurally and developmentally distinct. In contrast, bracts (leaves co-opted into floral function) are modular relative to true leaves; i.e., they maintain phenotypic stability in the face of environmentally induced leaf variation.

S23-04 Looking for modularity in morphometric data: Exploratory search without a hypothesis

Klingenberg, Chris (University of Manchester, GBR)

Modularity is a key concept in evolutionary developmental biology, where it is applied in a wide range of contexts from gene regulation to morphology. Many morphometric studies of modularity have used a-priori hypotheses derived from biological reasoning. Nevertheless, it often happens that no hypothesis of modularity is available, so that it is necessary to conduct an exploratory search for modules. Previous studies of this kind have used various clustering methods to find modules in morphometric data. This talk will evaluate the advantages and shortcomings of those methods and present a combination of methods that can be used in the search for modules. No single method is likely to provide satisfactory answers, but an approach combining multiple aspects is more promising.

14.00 – 15.40 Symposium S24: Origin and diversification of regeneration

ROOM C1

Organizer: Florian Raible
Chair: Florian Raible

S24-01 Taxon-specificity in Salamander limb development and regeneration

Brockes, Jeremy (University College London, GBR)

Salamanders are the only adult tetrapod vertebrates able to regenerate their limbs. Limb regeneration could either be an ancestral vertebrate property that is lost by other tetrapods, or could reflect some evolutionary novelty in salamanders. From an Evo-Devo perspective, limb development in larval salamanders is different from that in other tetrapods, particularly in relation to the property referred to as pre-axial dominance (PAD). It has been suggested that the origins of PAD, which are reflected in the fossil record of the Paleozoic branchiosaurid Apateon, may be connected to the origin of limb regeneration. It is unclear how they might be connected, and whether PAD operates in limb regeneration in extant species. We have described the salamander-specific three-finger protein called Prod 1, which was identified and analysed for its role in newt limb regeneration. Prod 1, which is readily identified as a unique structure by its twelve residue alpha-helical stretch, is not found in other vertebrate genomes such as zebrafish or Xenopus. We have analysed the expression and functional role of Prod 1 in larval forelimb development in the newt, using TALEN-mediated gene disruption. We also find that PAD is a feature of larval limb regeneration, in particular the early appearance of digits 1 and 2. Our work supports the idea that an evolutionary novelty that impacts on the development of the larval salamander limb, may also impact on its ability to regenerate this structure.

S24-02 Planarians as model system for the evolution of regeneration

Rink, Jochen (Max Planck Institute for Molecular Cell Biology and Genetics, Dresden, GER)

Some animals have astonishing regenerative abilities, including the regrowth of entire body parts lost to injury. However, already closely related species are often incapable of regeneration. Why in a world shaped by "survival of the fittest" regeneration should remain the exception rather than the rule remains one of the big questions in biology. Addressing this problem necessitates (1) some understanding of how regeneration works in the first place; (2) insight into what goes wrong in regeneration-deficient relatives; and (3) examination of the evolutionary pressures acting on regeneration as a selectable trait. Planarian flatworms provide an ideal system for addressing all three facets of the problem. These triploblastic animals are famous for their regenerative abilities and research over the past decade has identified some of the mechanisms that guide the regrowth of planarian tissue pieces back into a perfectly proportioned animal. We and others have shown that Wnt signaling plays a pivotal role in the maintenance and regeneration of the planarian body plan. High levels signify "tail fate" while inhibition of the pathway is necessary and sufficient for head specification. However, not all of the many hundred planarian species worldwide regenerate. Species exist that are either partially or even entirely regeneration deficient, thus providing unique opportunities for comparative regeneration research. As proof of principle, we took into the lab the European species *Dendrocoelum lacteum* that cannot regenerate a head from wound sites in the tail half of the animal. RNAi-mediated inhibition of Wnt signaling completely rescued the phenotype, thus illustrating that the miss-regulation of a signaling pathway can serve both as cause and cure of a species-specific regeneration defect. My lab is now in the process of establishing a large live collection of planarian species at MPI-CBG. The ~40 species that we have currently

at hand and further field sampling will allow us to systematically investigate the hypothesis that trade-offs between regeneration and sexual reproduction ultimately determine why some worms regenerate while others do not.

S24-03 Germline replacement in the Crustacean, *Parhyale hawaiensis*

Patel, Nipam (University of California Berkeley, CA, USA)

Studies in model species have revealed many of the genetic networks that guide development, and have opened the door to understanding how evolutionary changes in these networks lead to morphological and developmental diversity. I will describe our recent studies to understand developmental variation, focusing on the germline in the crustacean, *Parhyale hawaiensis*. *Parhyale* derives its primordial germ cells from a single precursor cell at the eight-cell stage. If this cell is ablated, the animal hatches without a detectable germline, but remarkably these animals are fertile as adults. We have been able to determine the source and timing of this replacement and are characterizing the molecular basis of this phenomenon.

S24-04 Injury-induced cell death, an evolutionarily-conserved force to drive regeneration?

Galliot, Brigitte (University of Geneva, CHE); Reiter, Silke (CHE); Wenger, Ivan (CHE); Chara, Osvaldo (CHE); Martinvalet, Denis (CHE); Buzgariu, Wanda (CHE)

To study how exogenous damages in an adult organism induce an injury response that is translated into wound healing, tissue repair, or 3D reconstruction, we use the freshwater cnidarian *Hydra* as a model system. *Hydra* is a tube-shaped animal with a head at the apical extremity (mouth, tentacles) and a basal disk (foot) at the basal one, which posseses the amazing ability to regenerate any missing part upon bisection of its body column. Hence it is possible to compare in the same animal 1) wound healing, 2) head regeneration, a complex morphogenetic process relying on the rapid local reprogramming of the gastric tissue into an organizer, and 3) foot regeneration, a process close to tissue repair. This tissue plasticity is possible thanks to the three stem cell populations that constantly self-renew in *Hydra*. Studies from our laboratory identified an immediate activation of the MAPK/CREB pathway in head regenerating tips after mid-gastric bisection, which leads to a wave of apoptosis over the first hours of head regeneration (Kaloulis et al. 2004; Chera et al. 2009, 2011). Apoptotic cells transiently release Wnt3, which activates the b-catenin pathway in the surrounding progenitors, leading to the formation of a proliferative zone, a process similar to apoptosis-induced compensatory proliferation in Drosophila wing discs. Interestingly this process is not observed in simple wounds and is asymmetrical, i.e., present in head-regenerating tips, but not in foot-regenerating ones. However, ectopic activation of cell death in foot-regenerating tips suffices to trigger head regeneration. We will present recent results on the signals released at the wound surface that possibly trigger cell death, and discuss whether cell death can be considered as an evolutionarily conserved process to drive regeneration across animal species.

14.00 – 15.40 Symposium S25: "Living fossils", myth or reality?

ROOM C2

Organizers: Patrick Laurenti and Didier Casane
Chairs: Didier Casane and Patrick Laurenti

S25-01 "Living fossils" the facts beyond the myth

Laurenti, Patrick (CNRS, Université Paris-Diderot, Sorbonne Paris Cité, Gif-sur-Yvette, FRA); Casane, Didier (CNRS, Université Paris-Diderot, Sorbonne Paris Cité, Gif-sur-Yvette, FRA)

"Living fossils", a term first coined by Darwin, have been used to characterize extant species that have no (or few) close living relatives and that were thought to undergo morphological stasis over long periods of evolutionary time. Recently, the genomes of several vertebrates considered as "living fossils" or "ancient lineages" were shown to have a low substitution rate in protein coding sequences. This finding led to a controversy, with some arguing that slow genome evolution is correlated to morphological stasis, whereas others argue that morphological evolution is not correlated to molecular evolution in such a simplistic way. Evolutionary genetics principles state that a low molecular evolution implies either a low mutation rate and/or a strong selection against new alleles. Therefore, the identification of bona fide slow evolving genomes may reveal an unusual high accuracy of DNA replication (or very efficient DNA repair mechanisms), and/or an unusually strong purifying selection. We will show that vertebrate species that display a low substitution rate in coding sequences do not have an overall slow-evolving genome: these species can display genomic rearrangements, and/or active transpositions, and/or a "normal" substitution rate at neutral sites. In addition, not only morphological evolution and molecular evolution are not correlated simply, but also vertebrate species that are often presented as "ancient", "primitive", or "ancestral" actually display many derived traits. We will therefore question the role of the low substitution rate in morphological stasis.

S25-02 Coelacanth and the myth of living fossil

Dutel, Hugo (RIKEN Center for Developmental Biology, Kobe, Hyogo, JPN)

Coelacanths (Actinistia) are a group of lobe-finned vertebrates (sarcopterygian fishes) that have been described in the fossil record since the 19th century. Because the last occurrences of fossil coelacanths are

late Cretaceous in age, the group was thought to be extinct at the end of this period (about 70 million years ago). As such, the discovery of a living coelacanth offshore South Africa in 1938 has raised great excitement among the scientific community. The living coelacanth *Latimeria chalumnae* was even more sensational since its morphology appeared to be very similar to those of its extinct relatives. As a result, *Latimeria* was rapidly popularized as a "living fossil". Moreover, coelacanths were seen at this time as the closest relatives of tetrapods, and *Latimeria* as the direct "descendant" of a group of sarcopterygian fishes (rhipidistians) that was supposed to be at the origin of dwelling vertebrates (tetrapods). In this context, the newly discovered living coelacanth *Latimeria* was expected to allow a better understanding of the biology of rhipidistian fishes, and of the fish-tetrapod transition during the Devonian. Since then, the phylogenetic analyses of morphological, molecular, and, more recently, whole genome data have shown that coelacanths are more distantly related to tetrapods than previously thought. However, the concept of "living fossil" has remained tightly associated with coelacanths despite of the use of tree-thinking approaches (involving mosaic evolution concept) to investigate their intra- and inter-relationships. Based on comparative anatomy and morphology, it is clear that the long-standing idea of coelacanths as living fossils (notwithstanding the oxymoron itself) is specious and wrong. We will show how paleontology questions the traditional view of coelacanths as a group that has experienced little anatomical and environmental changes throughout its evolutionary history. Then, we will present recent developmental data acquired on *Latimeria* using x-ray synchrotron microtomography, which emphasis the derived features of *Latimeria*, and suggest that developmental heterochronies have played a key role in the early evolution of the coelacanth anatomy. Finally, we will present how the better understanding of the functional morphology and ecology of *Latimeria* is expected to give new insights in the constraints and trade-off that accompanied coelacanth and lobe-finned fishes morphological evolution.

S25-03 Evolution and diversity of the Chondrichthyes

Cuny, Gilles (Natural History Museum of Denmark, Copenhagen, DNK)

With their cartilaginous skeleton, sharks, skates, rays and chimaeras are often considered as the most "primitive" gnathostomes on Earth, and as such are often used in phylogenetic, molecular or not, studies as outgroups and proxies for the ancestral state of many characters. New fossil discoveries, like for example the placoderm *Entelognathus*, strongly suggest, however, that their cartilaginous skeleton is in fact a reversion, and that the simplistic evolutionary grading of gnathostomes: Placoderm, chondrichthyan, acanthodian and finally osteichthyan is indeed wrong. Osteichthyans are likely to be the sister-group of the placoderms, while chondrichthyans+acanthodians represent a separate, more derived clade. The typical sharky shape of the modern selachimorphs is also often seen as a primitive pattern that did not evolve much through the evolution of the group. This is to forget that this typical shape evolved several time independently, in the Devonian *Cladoselache* and in the modern selachimorphs for example. Its design also shows some major improvements, like for example the appearance of calcified vertebral centra in the selachimorphs around the Permian/Triassic. The emblematic teeth of the sharks are also often seen as a primitive feature that did not evolve much during their evolution, but again their structure, more particularly the enamel-like tissue making their external cover went through a lot of improvements through time, even if recent discoveries make the deciphering of the details of the processes a little more challenging than we used to think.

S25-04 Embryology of the hagfish and early evolution of vertebrates

Kuratani, Shigeru (RIKEN Center for Developmental Biology, Kobe, Hyogo, JPN), Ota, Kinya G. (Marine Research Station, Yilan, TWN), Oisi, Yasuhiro (Max Planck Florida Institute for Neuroscience, Jupiter, FL, USA)

Living jawless veretebrates consist of hagfish and lamprey, and the monophyly of this group is now well established by recent molecular analyses. Comparative morphology of these two cyclostomes, however, has contradicted the monophyly, especially for the apparent lack of some vertebrate-defining traits, like vertebrae, in the hagfish: the morphology of hagfish has been interpreted to represent a more basal animal than that of the rest of vertebrates. In addition, the hagfish adenohypophysis and related structures were suggested to arise from endoderm, unlike that of other vertebrates derived from the ectoderm: reexamination of hagfish embryology is critical to evaluate the anatomical traits of cyclostomes. By observing staged hagfish embryos, we show that the hagfish adenohypophysis arises ectodermally, as a posterior part of the medial placode, the hypophyseal plate, as in the lamprey larva. This finding allowed us to identify a craniofacial developmental pattern common to cyclostomes, but not to crown gnathostomes. From this cyclostome-specific developmental stage, lamprey and hagfish develop into distinct developmental trajectories, making it difficult to establish morphological homologies in adult anatomy of these animals. We also show that the comparison with gnathostomes, the out-group of cyclostomes, implies that many of the hagfish peculiarities can be recognized as hagfish-specific derived traits (autoapomorphies). Thus the lamprey is likely to represent more ancestral state of cyclostomes, possibly reflecting the morphological and developmental pattern of the latest common ancestor of entire vertebrates. Based on the above developmental scheme, we first showed homologies of skeletal elements between lamprey and hagfish chondrocrania.

16.10 – 17.10 **Contributed Session C25:**
'NEPTUNE' ITN: The evolution of sensory systems in the marine environment

ROOM A

Chair: Gaspar Jékely

C25-01 An ancient neuropeptide regulates both larval settlement and feeding in the marine worm *Platynereis dumerilii*

Williams, Elizabeth (Max Planck Institute for Developmental Biology, Tübingen, GER); Conzelmann, Markus (Max Planck Institute for Developmental Biology, Tübingen, GER); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER)

During larval settlement and metamorphosis, marine invertebrates undergo changes in habitat, morphology, behavior and physiology. How larvae co-ordinately regulate these changes is not well understood. Recently, we discovered a neuropeptide, myoinhibitory peptide (MIP), as an important regulator of larval settlement behavior in the marine annelid Platynereis dumerilii. MIP is a member of the ancient eumetazoan Wamide neuropeptide superfamily that includes insect allatostatin B and Cnidarian GLWamides and is associated with life cycle transitions in many animal phyla. Here, we explore the contribution of MIP to feeding, another aspect of Platynereis life cycle progression. We find that MIP is expressed in both the brain and gut of developing Platynereis larvae. Activating MIP signaling by synthetic neuropeptide addition causes increased gut peristalsis and pharynx extension. Conversely, morpholino knockdown of MIP expression delays the onset of feeding in Platynereis larvae. These results indicate that additional to its role in settlement, MIP is a regulator of Platynereis late larval and juvenile feeding and digestion. The dual roles of MIP in feeding and settlement may promote the coupling of these two processes during life history evolution. Through our integrative analysis of MIP function, we begin to establish Platynereis as a powerful model for studying the neuroendocrine regulation of feeding.

C25-02 Regulation of apical sense organ formation in the sea anemone *Nematostella vectensis* by Frizzled 5/8 and Glypican 4/6

Bause, Markus (University of Bergen, NOR); Rentzsch, Fabian (University of Bergen, NOR); Leclère, Lucas (Observatoire Océanologique de Villefranche sur Mer, FRA); Sinigaglia, Chiara (Observatoire Océanologique de Villefranche sur Mer, FRA)

The apical organ is considered to be the major larval sense organ in some cnidarian & other marine larvae (including protostomes & deuterostomes). It is always at the pole opposite to the blastopore and it is characterized by a long ciliary tuft. The relationship of apical organs between the phyla remains unclear. The apical organ of *Nematostella vectensis* has a major function in triggering metamorphosis and it has been proposed to possess light-, chemo- and mechanoreception. Our study focuses on the molecular mechanisms that govern apical organ development and we show here that the Wnt receptor Frizzled5/8 and Glypican4/6 are two essential regulators of the development of the apical territory. Knockdown of *frizzled 5/8* results in a shortened oral-aboral axis and a shift of expression patterns along that axis. Glypicans can function as co-receptors in almost all the major developmental signaling pathways, including the Wnt and FGF signalling. Knockdown of *glypican4/6* leads to a similar phenotype than *frizzled5/8* knockdown at gastrula. However, in contrast to knockdown of *frizzled 5/8*, but similar to knockdown of *FGFa1*, *glypican4/6* morphants do not initiate apical organ formation after gastrulation. These results suggest that Glypican 4/6 functions in Frizzled 5/8 mediated patterning of the apical territory before gastrulation and is possibly involved in FGF mediated apical organ specification after gastrulation.

C25-03 Expression of neuropeptides in the brachiopod, *Terebratalia transversa*

Thiel, Daniel (University of Bergen, NOR); Jékely, Gáspár (Max Planck Institute for Developmental Biology, Tübingen, GER); Hejnol, Andreas (University of Bergen, NOR)

Animal nervous systems utilize a wide array of neuropeptides and the study of these can provide insight into the architecture, function and evolution of nervous systems. Contrary to other neurotransmitters such as acetylcholine or serotonin, which mainly transfer action potentials between neurons, neuropeptides are released slower and are often involved in triggering physiological, developmental and behavioral events such as feeding, reproduction, settlement and metamorphosis. Up until now, nothing is known about the neuropeptides in brachiopods, an understudied group of the Trochozoa. Using comparative genomics and transcriptomics, we identified 14 neuropeptides that are expressed in the nervous system of the brachiopod, *Terebratalia transversa*. Some of these neuropeptides are present in a wide range of metazoans, (e.g., 7B2, insulin-related peptides, vasotocin-neurophysin, glycoprotein hormones). Others are more restricted to protostome lineages, with several belonging to groups with conserved amidated C-terminal amino acid sequences, like YRLamide (RLamides are known from other Trochozoans as well as from Arthropods), RGWamide (GWamides are known from annelids, molluscs, platyhelminths and arthropods) and FLRFamide (FLRFamide is known from molluscs and annelids and with N-terminal extension also from some other Protostomia, while RFamides in general can be found throughout metazoa). We investigated their expression pattern in embryonic and larval stages by using immunohistochemistry and in situ hybridization. Nearly all of them are

expressed in the apical lobe of early and late larvae. Surprisingly a subset of these neuropeptides is already expressed during gastrula stages. The comparision of the neuropeptide complement and expression in brachiopods with other animals will contribute to a better understanding of nervous system evolution.

C25-04 Evolution and development of photoreceptors in Polyplacophora

Vöcking, Oliver (University of Bergen, NOR); Hausen, Harald (University of Bergen, NOR)

Invertebrate light sensing organs show a great variety in structure and function leading to the questions how this diversity evolved. Several kinds are described in mollusks from simple to complex cerebral eyes and various extraocular photoreceptors, but more detailed studies refer exclusively to representatives of conchiferans like cephalopods, gastropods or bivalves, which all have a well developed ganglionated nervous system. We chose a phylogenetically distinct polyplacophoran representing those molluscs with a simpler tetraneural type nervous system as a study object (1) to obtain insights in the mollusk ancestral set of photoreceptors and (2) since homology of polyplacophoran cerebral eyes is a matter of debate due to their unusual position and developmental origin. Here we characterize different photoreceptors of the chiton *Leptochiton asellus* by genes patterning the larval neuroectoderm, eye and CNS developmental genes and effectors involved in photoreceptor physiology. The data provide new insights into the evolutionary origin of chiton eyes, but also on the mechanism of photopigment renewal in mollusks and beyond. Beside in the eyes, we also found and characterized for the first time photoreceptors in the apical organ of a mollusk providing further evidence that light sensation is an important feature of this main larval neuronal structure.

16.10 – 17.10 Contributed Session C26: Quantitative EvoDevo in model and non-model organisms II

ROOM B

Chair: Philipp Mitteroecker

C26-01 A differentially expressed gene network in the head of divergent arctic charr morphs is potentially regulated by Ets-2

Pashay Ahi, Ehsan (University of Iceland, Reykjavik, ISL); Hristova Kapralova, Kalina (University of Iceland, Reykjavik, ISL); Pálsson, Arnar (University of Iceland, Reykjavik, ISL); Helene Maier, Valerie (University of Iceland, Reykjavik, ISL); Gudbrandsson, Jóhannes (University of Iceland, Reykjavik, ISL); Snorrason, Sigurður (University of Iceland, Reykjavik, ISL); Franzdóttir, Sigrídur R. (University of Iceland, Reykjavik, ISL); Jónsson, Zophonías O. (University of Iceland, Reykjavik, ISL)

The extensive craniofacial diversity in teleost fish provides an interesting model for studying structural variations that arise during development and play a role in evolution. Arctic charr, *Salvelinus alpinus*, is one of several northern freshwater fish species exhibiting parallel evolution relating to benthic versus limnetic ecology and often reflected in morphological variation, particularly in the head and trophic apparatus. In Lake Thingvallavatn, four morphs of Arctic charr are found; two benthic morphs with subterminal mouth and blunt snout (small and large benthivorous charr) and two limnetic morphs with a terminal mouth (planktivorous and piscivorous charr). The distinct craniofacial variation is observed upon hatching. We have profiled the transcriptome of two contrasting morphs at three stages prior to hatching using RNA-seq thereby generating a list of genes that are differentially expressed among morphs. In this study, a combination of gene expression and bioinformatic tools was used to confirm and characterize differentially expressed genes related to craniofacial morphogenesis. Among these, we identified a network of genes displaying conserved co-expression across vertebrates. Many of these genes are involved in extracellular matrix remodelling and intramembranous ossification. Moreover, analysis of conserved regulatory regions of the co-expression network genes across three fish species showed conserved binding sites for several transcription factors, including Ets-2 and Ap-1. Ets-2 itself was found to be differentially expressed between benthic and limnetic morphs and its expression level correlated positively with expression of the network genes. Furthermore, a substantial spatiotemporal overlap (in the lower jaw and pharyngeal arches) was seen in mRNA expression of Ets-2 and other genes of the network. Thus, we hypothesize that a mechanism for the differential regulation of this gene network, possibly through the transcription factor Ets-2, may play an important role in benthic/limnetic craniofacial divergence of Arctic charr morphs in this lake.

C26-02 Gap domain shifts caused by damped oscillations represent a dynamic fossil of short-germband evolution

Verd, Berta (Center for Genomic Regulation, Barcelona, ESP); Crombach, Anton (Center for Genomic Regulation, Barcelona, ESP); Jaeger, Johannes (Center for Genomic Regulation, Barcelona, ESP)

Insects use two main modes of segmentation during development: the more ancestral short-germband mode (e.g., *Gryllus bimaculatus*), where new segments are periodically produced from a posterior growth zone, and the long-germband mode (e.g., *Drosophila melanogaster*), where body segments are formed by simultaneous subdivision of the embryo. In dipterans (flies, midges, and mosquitos), which use the long-germband mode of segmentation, the gap genes are activated by maternal gradients and will cross regulate each other to form the first zygotic regulatory layer of the segmentation gene hierarchy. Using

quantitative spatio-temporal expression data we have been able to obtain a very precise mathematical model of the gap genes in *D. melanogaster*, with which to study the dynamics of pattern formation. This model has made clear that distinct dynamical regimes are responsible for shifting and non-shifting gap gene domains and that, in fact, the shifting observed in the posterior trunk domains is driven by a damped oscillator. This is the first instance of an oscillator of any sort found to be involved in the segmentation process of a long-germband insect. We have been able to identify a three-gene motif embedded in the underlying gap gene regulatory network as enough to explain both shifting and non-shifting domains. One or another dynamical regime will govern the dynamics of gene expression depending on which gap genes are involved. This motif, known as the AC/DC circuit, can exhibit one added dynamical regime: it can drive oscillations. This means that the motif responsible for the dynamics of trunk gap gene patterning in a long-germband insect oscillates under certain conditions, and oscillations are characteristic of short-germband segmentation. This finding connects both segmentation modes suggesting that they might be significantly more similar than previously thought. Our work therefore suggests that the damped oscillation-based mechanism driving patterning in the posterior trunk region of the *D. melanogaster* embryo is a dynamical fossil representing a signature of the ancestral, short germband mode of segmentation. This result can help us better understand the evolution of the various segmentation modes in insects.

C26-03 Quantification of developmental variation in rainbow trout using geometric morphometric image analysis

Mayer, Christine (University of Vienna, AUT); Metscher, Brian (University of Vienna, AUT); Müller, Gerd B. (University of Vienna, AUT); Mitteroecker, Philipp (University of Vienna, AUT)

The ways in which embryo development can vary across individuals of a population determine how genetic variation translates into adult phenotypic variation. Progress in studying developmental variation is impeded by the lack of quantitative methods for analyzing the geometry of developing embryos in concert with the spatial patterns of cellular activity. By drawing on the strengths of geometric morphometrics and image analysis, we present a new approach for the statistical analysis of embryonic images. The shape of well-differentiated embryonic structures is parameterized by landmarks and semilandmarks, whereas the spatial pattern of cell density is described by the registered images. We apply this approach to microscopic images of the tail fins of juvenile rainbow trout. We were able to demonstrate how average fin shape and average cell density change within a period of 35 days, including the emergence of the fin rays as a novel tissue structure. Inter-individual variation of tissue density is highly spatially structured and temporally dynamic throughout the investigated period.

C26-04 Compartmentalization and spatial complexity of gene expression through development

Salvador-Martínez, Irepan (University of Helsinki, FIN); Salazar-Ciudad, Isaac (University of Helsinki, FIN)

During animal development, an apparent homogeneous cell transforms into an organism with many cell types arranged in specific spatial patterns. This process has been described as the progressive compartmentalization of the embryo. At the expression level, it is assumed that genes start being expressed in wide areas of the embryo and as development proceeds, they tend to be expressed progressively in a more spatially restricted and complex areas. Although there is qualitative knowledge about this process, up to this point there has not been any systematic quantitatively attempt to measure if that is the case, or if it happens in the same way in different species and developmental stages. Here we present a morphometric quantitative analysis of the compartmentalization and spatial complexity of gene expression in three distantly related species *(Drosophila melanogaster, Xenopus leavis, Ciona intestinalis*) over developmental time. For this analysis, we analyzed thousands of gene expression spatial patterns from in situ hybridization databases. Our results provide a quantitative evaluation of the widespread view that compartmentalization and complexity increase during development. Spatial complexity (understood as a "roughness" measure of the outline of expression) not only increases progressively but also do it at a progressively finer-grained scale with time. We found some differences between the species. *Drosophila* displays the earliest and most sudden change in both compartmentalization and complexity, just before or at the beginning of gastrulation. In *Ciona* these changes were less sudden and after gastrulation. In *Xenopus*, the major changes were in the tailbud stages. Additionally, in all species transcription and growth factors showed an early compartmentalization (compared to other genes), which is consistent with those genes being more directly involved in early pattern formation, and transcription factors decrease their area of expression faster than other genes. Finally, for each stage we measured the number of cells with a unique combination of expressed genes. Interestingly, until the 32-cell stage the *Ciona* embryo is totally compartmentalized (every blastomere has a unique combination of genes expressed), and from that stage the number of compartments increases but the embryo becomes less compartmentalized compared to its cell number (many cells are equivalent and expresses the same sets of genes).

16.10 – 17.10 **Contributed Session C27:**
How does developmental robustness facilitate the evolution of biodiversity?

ROOM C1

Chairs: Günter Theissen and Rainer Melzer

C27-01 The significance of developmental robustness for the biodiversity of life

Melzer, Rainer (Friedrich Schiller University Jena, GER); Theissen, Günter (Friedrich Schiller University Jena, GER)

The causes of the origin of species biodiversity are still poorly understood. In both plants and animals, the vast amount of species diversity is produced by only a few taxonomic groups. Intriguingly, these species-rich taxa often have highly standardized body plans. This is rather counter-intuitive as one may initially assume that morphological diversity and species diversity are positively correlated with each other. Another way to explain the success of certain taxa over others is the origin of evolutionary novelties. Indeed, there is no doubt that key innovations profoundly contributed to evolutionary success. However, they cannot entirely explain why some taxa have been more successful than others. For example, the key innovations associated with the angiosperm flower, i.e., the carpel, bisexuality and petals are all already present in early diverging angiosperms. Nevertheless, early diverging angiosperms are by far less species-rich than many of the later branching lineages. We speculate that one of the major trends in plant and animal evolution is an increase in developmental robustness. Whereas the early diverging lineages of a certain taxonomic group already possess a characteristic evolutionary novelty, the phenotypic manifestation of this novelty is not yet very robust. Thus, although evolutionary innovations may have set the stage for the success of certain taxa, processes that conferred robust developmental control over these key innovations might have been equally important. Taken together, an increased level of robustness may foster species diversity and may at the same time limit morphological diversity.

C27-02 A computational approach to the evolution of development under conservative selection

Zimm, Roland (University of Helsinki, FIN); Salazar-Ciudad, Isaac (University of Helsinki, FIN)

The velocity of morphological and developmental change during evolution differs between clades and is believed to be affected by the strength of external forces driving evolution, such as disruptive selection. However, even under conservative selection pressures, i.e., the favouring of morphological invariance, development can diverge between closely related and phenotypically similar species. Understanding the rules that underly these changes is relevant both for understanding the evolution of development and morphological variability and, potentially, the origin of evolutionary innovations. We hypothesize that the way these pattern transformations are arranged between each other during developmental time affects the ways in which development as a whole can evolve. We built a general model of development (with gene networks, extracellular signal diffusion, cell behaviors such as cell division, apoptosis, adhesion, contraction, extracellular matrix secretion, etc.) and put it to evolve by mutation under conservative selection in a population, i.e., running the developmental model under the different mutated genotypes gives rise to different phenotypes. This setup allows us to arrange the pattern transformations occurring during development of the founder populations in several ways, e.g, whether signalling and cell movements occur simultaneously (morphodynamic) or sequentially (morphostatic) and when during development either of these types predominate. We observe in our simulations of evolution that in many cases morphodynamic mechanisms get replaced by morphostatic ones over time, since these are more likely to produce the same phenotype under conservative selection. Second, we found that within evolutionary time, network interconnectivity increases due to developmental systems drift, which makes mutations increasingly likely to have some impact on the phenotype and be less likely selected. Consequently, changes tend to occur rather early during the course of artificial evolution. Furthermore, we observe that early stages of development change less often in our simulations, which is particularly the case in extensively morphostatic developments. This is consistent with von Baer's law according to which phenotypic variation increases steadily during development.

C27-03 Divergent role of the Hox gene *Antennapedia* in spiders is responsible for the convergent evolution of abdominal limb repression

Khadjeh, Sara (University of Göttingen, GER); **Turetzek, Natascha** (University of Göttingen, GER); Prpic-Schäper, Nikola-Michael (University of Göttingen, GER)

Similar bodyplan features are found in very distantly related groups, for example the leg free "abdomen" of spider and flies. This body region has evolved convergently in these groups. Hox genes serve as master regulators for segment identity and thus also control the development of body regions. The abdomen of insects is controlled by the posterior Hox genes *Ubx* and *abd A*. Spiders also have a leg free posterior body tagma, the opisthosoma. Interestingly we found that *Antp* from the spider *P. tepidariorum* is expressed in the opisthosoma and parental RNAi knockdown results in the development of an extra leg pair on the first opisthosomal segment. To test whether the remaining opisthosomal Hox genes have a redundant function in leg repression we are establishing methods to knock down several Hox genes simultaneous-

ly. Preliminary results of double pRNAi of *Pt-Antp* and *Ubx* result in the development of a paired appendage rudiment on the second opisthosomal segment. In addition, we found that *Ubx* from another spider species, *Pholcus phalangioides,* is only expressed in one opisthosomal segment and thus might have changed its leg repressive function due to its possible redundancy with *Antp*. To test whether the functional change of *Antp* in spiders and flies is caused by the protein sequence itself, we ectopically expressed *Pt-Antp* in the eye-antenna imaginal disc of *D. melanogaster*. The results show the same antenna-to-leg transformation as known for *Dm-Antp*, suggesting that the functional change is caused by divergent evolution of cofactors, rather than the protein itself. *homothorax (hth)* and *extradenticle (exd)*, are important Hox co-factors and ubiquitously expressed in the wild type antennal imaginal disc. The misexpression of *Pt-Antp* and *Dm-Antp* in antennal imaginal discs resulted in either downregulation of *hth* or *exd*, respectively . This regulatory switch may provide insights into the evolution of Hox co-factor regulation and the principles of convergent evolution.

C27-04 Designing a mesodermal molecular toolkit in the marine annelids *Alitta virens* and *Platynereis dumerilii*

Kozin, Vitaly V. (St. Petersburg State University, RUS); Raible, Florian (Max F. Perutz Laboratories, University of Vienna, AUT); Kostyuchenko, Roman P. (St. Petersburg State University, RUS)

Germ layers are profound features of animal development. They originate in an early embryogenesis and predetermine the future fates of extensive cell masses. During the further process of cell differentiation, each germ layer gives rise to a similar complement of derivatives in diverse metazoans. Mesoderm generally produces muscles, coelomic epithelium and connective tissues, but even among bilaterians, a distinct cell type, striated muscles for instance, may share only a limited core molecular apparatus, differing in essential functional elements (Steinmetz et al. 2012). This raises the question to which extent the developmental programs that drive germ layers specification and differentiation are conserved. Representatives of the group Spiralia (flatworms, annelids, and molluscs) are known for extremely stereotyped patterns of early development. As a rule, one and the same cell — 4d — gives rise to the whole trunk mesoderm, facilitating investigation of this germ layer and comparisons among the group. Polychaete worms display a compound segmented body plan similar to the hypothesized Urbilateria. Molecularly, an ancestral-type genome organization has been shown for the nereid polychaete, *P. dumerilii* (Raible et al. 2005). Additional data suggest that both the developmental programs and the molecular anatomy of nereid polychaetes have ancient-type characteristics (Denes et al. 2007; Christodoulou et al. 2010; Simakov et al. 2013). Along with the early Cambrian origin of these animals, this makes them particularly attractive for Evo-Devo studies. In the present work we focused on the mesoderm development of the two marine annelids *A. virens* and *P. dumerilii*. Expression analyses of selected molecular markers revealed mesodermal expression of *twist, mox, vasa, pl10* and *piwi* genes, whereas *evx* and *gcm* were restricted to ectodermal derivatives. Furthermore, we studied a possible role of MAP kinase signaling in early mesoderm specification by applying the MAPKK inhibitor U0126 during different cleavage stages. U0126 prevented formation of functional muscles, and disrupted the normal metameric *twist* and *mox* patterns, indicating a role of the MAPK cascade for proper morphogenesis and differentiation of mesodermal tissues. To further dissect the molecular toolkit of mesoderm development, we are taking additional steps towards the generation of transgenic and knock out animals.

16.10 – 17.10 Contributed Session C28: Uncovering the genomic bases of phenotypic change in the NGS era II

ROOM C2

Chairs: Juan Pascual and Ignacio Maeso

C28-01 Evolution of the eye transcriptome under constant darkness in Sinocyclocheilus cavefish

Meng, Fanwei (Chinese Academy of Sciences, Beijing, CHN); Braasch, Ingo (University of Oregon, Eugene, OR, USA); Phillips, Jennifer (University of Oregon, Eugene, OR, USA); Zhang, Chunguang (Chinese Academy of Sciences, Beijing, CHN); Postlethwait, John (University of Oregon, Eugene, OR, USA)

Cave animals evolve distinct troglomorphic characters in perpetual darkness, most commonly, the reduction of eyesight, pigmentation, pineal organ, and scales, countered by augmented chemoreceptors, mechanoreceptors, and lipid storage. In cave *Astyanax mexicanus*, lens apoptosis associated with over-expression of *sonic hedgehog* induces eye degeneration. The freshwater teleost genus *Sinocyclocheilus* is endemic to the southwestern China. The genus includes many surface species and at least 10 cave species with different degrees of eye degeneration. The research on a variety of cave-animal models is necessary to understand whether independent evolutionary lineages utilize related molecular genetic mechanisms.

C28-02 Evolution of the head developmental gene regulatory network in three closely related Drosophila species

Torres Oliva, Montserrat (Georg August University of Göttingen, GER); Almudi, Isabel (Oxford Brookes University, GBR); Nunes, Maria D. S. (Oxford Brookes University, GBR); McGregor, Alistair P. (Oxford Brookes University, GBR); **Posnien, Nico** (Georg August University of Göttingen, GER)

The size and shape of an organism and its organs has to be tightly controlled during embryonic and postembryonic development to ensure proper functionality. However, these adult features are certainly target for evolutionary changes leading to the breath-taking diversity of body forms observable in nature. This contradiction suggests that developmental gene regulatory networks (GRNs) are constrained to a certain level, but nodes within this network are prone to change to give rise to morphological divergence. One aim of our research is identifying flexible nodes within the GRN underlying adult head formation in Drosophila. We previously showed that head shape and compound eye size varies considerably within and between the three sibling species *D. melanogaster, D. simulans* and *D. mauritiana*. The difference in eye size is mainly due to a combination of variation in ommatidia number and size. We use the development of this complex adult structure as a system to study variation on a GRN that overall should be very similar, but obviously has evolved to produce significant size and shape differences. To this aim we apply RNAseq to unravel the core transitions in the GRN during eye-antennal imaginal disc development. Subsequently, we identify those genes differentially expressed between species with morphological differences to find candidate nodes of the network prone to evolve. This search for the varying nodes is aided by quantitative trait loci (QTL) mapping what allows a more rapid identification of candidate genes. The results obtained will be discussed in the context of GRN constraints and flexibility.

C28-03 Evolutionary innovation by rewiring of gene networks: Origin of sense organs in the vertebrate "new head"

Riddiford, Nick (National University of Ireland, Galway, IRL); Schlosser, Gerhard (National University of Ireland, Galway, IRL)

How novel characters arise remains one of the most perplexing questions in evolutionary biology. To examine this we must explore how ancestral genetic networks have been rewired during evolution, and how this rewiring promoted the development of novel traits. We aim to elucidate the gene regulatory network (GRN) that underlies the formation of vertebrate cranial sense organs, and compare the revealed network with a reconstructed ancestral state. We use *Xenopus laevis* as a model for extant vertebrates, and will estimate the ancestral state through network reconstruction in *Ciona intestinalis* (which lacks homologues of these cranial sense organs). Expression of the genes *Six1* and *Eya1* marks the area of the embryo (pan-placodal region) that will later differentiate into the cranial sense organs. Mutation or knockdown of these genes blocks the development of normal sense organs and cranial ganglia by interfering with cell proliferation, neuronal differentiation and cell migration; thus, *Six1* and *Eya1* play central roles in the network underlying cranial sense organ development in vertebrates. To determine the GRN controlled by *Six1* and *Eya1* we have screened for their direct target genes in *Xenopus* by injecting embryos with hormone-inducible constructs of *Six1* and *Eya1*, which are hormone activated after treatment with the protein synthesis inhibitor cycloheximide. We have determined the complete transcriptome from placodes explanted from hormone treated embryos using RNA-Seq, and analysed differential gene expression using the Tuxedo bioinformatics suite. From the most differentially expressed genes we have selected putative target transcription factors that we are currently verifying by qPCR and in situ hybridisation. Subsequently, we will test for cross-regulatory interactions in gain and loss of function studies, and the network of interactions between homologous genes will be reconstructed through functional studies in *Ciona*. These studies, together with further gain and loss of function experiments in both *Ciona* and *Xenopus*, will allow us to elucidate evolutionary novelties in the gene network responsible for the development of vertebrate cranial sense organs.

C28-04 Expression and diversification of *CYCLOIDEA* genes in Asteraceae: A case from the highly derived tribe Anthemideae

Bello Gutiérrez, M. Angélica (Real Jardín Botánico, Madrid, ESP); Cubas, Pilar (Centro Nacional de Biotecnología, Madrid, ESP); Álvarez, Inés (Real Jardín Botánico, Madrid, ESP); Durán, Fátima (Real Jardín Botánico, Madrid, ESP); Sanjuanbenito, Guillermo (Real Jardín Botánico, Madrid, ESP); Fuertes Aguilar, Javier (Real Jardín Botánico, Madrid, ESP)

Floral symmetry is one of the most dynamic features of the flower. In the case of Asteraceae inflorescence, where several floral morphologies coexist in a capitulum, this evolutionary trait becomes more evident. Along several clades of Asteraceae, floral symmetry, shape and sexuality are extremely variable even within a single genus. In the highly evolved tribe of Anthemideae, a Mediterranean genus with 13 spp., Anacyclus, possesses heterogamous and homogamous inflorescences. The heterogamous are composed by bisexual, actinomorphic, pentamerous disc flowers and peripheral zygomorphic, trimerous, rayed female flowers. The species with homogamous capitula lack the peripheral, zygomorphic ray flowers. As *CYCLOIDEA (CYC)* genes are involved in the establishment and maintenance of differential floral

symmetry in eudicots, we explored the gene sequences and patterns of diversification of Anacyclus and other Asteraceae/eudicots using a phylogenetic approach. Bayesian analyses suggest that there are two *CYC1*, two *CYC3* and five *CYC2* genes in Anacyclus, a number of paralogs similar to those documented in Gerbera and Helianthus. Additionally, qPCR expression patterns of the Anacyclus *CYC2* genes were analyzed in vegetative organs, and disc, ray and naturally occurring ray variant flowers ("trumpet"). As observed in other Asteraceae there is a differential expression in ray vs disc flowers. Expression in AcCYC2.1, AcCYC2.2, AcCYC2.3, AcCYC2.4 is significatively increased in ray flowers as compared to disc flowers. The expression of these genes in trumpet flowers follows a similar pattern to that of disc flowers. In situ hybridization of *CYC2* probes (AcCYC2.1, AcCYC2.2, AcCYC2.4 and AcCYC2.5) were tested in order to detail tissue-specific expression at different stages of development. We have found contrasting patterns between different paralogs in ray and disc flowers. In disc flowers no expression of the *CYC2* probes was observed in the perianth. Expression persists in stamens and ovules from early to mid development. In mid stage-ray flowers the expression in ovules is already evident whereas it is barely perceptible in the abaxial perianth. Although qPCR and in situ hybridization resulted in differential expression of *CYC* genes in ray vs disc and trumpet flowers suggesting a role of *CYC* in the symmetry diversity in Asteraceae, in situ hybridization supports an additional *CYC* role in the development of stamens and ovules.

16.10 – 17.10

Contributed Session C29: Developmental mechanisms underlying evolutionary change III

ROOM D

Chair: Jack Green

C29-01 Evolution of the germ cells: Insights from a centipede

Green, Jack (University of Cambridge, GBR)

Germ cells are a unique cell type — the only cell population that makes a genetic contribution to the next generation. The comparison of germ cell specification and differentiation across the animal kingdom is emerging as a powerful study system to ask questions about how and why developmental mechanisms vary. By studying the centipede *Strigamia maritima* we provide the first systematic description of germ cell development with molecular markers in a myriapod. Through examining the expression of *Strigamia vasa* and *nanos* orthologues, we find that the primordial germ cells are specified from at least the blastoderm stage. This is a much earlier embryonic stage than previously described for centipedes, or any other member of the Myriapoda. Using these genes as markers we track the development of the germ cells throughout embryogenesis. We find that the germ cells accumulate at the blastopore; that the cells do not internalize through the hindgut, but rather through the closing blastopore; and that the cells undergo a long-range migration to the embryonic gonad. This is the first evidence for primordial germ cells displaying these behaviours in any myriapod. Finally, we also find a surprising localization of maternal Vasa protein within the germinal vesicle of developing oocytes, and propose a hypothesis for future work that this could act as mechanism for localizing Vasa protein asymmetrically in early embryos. The myriapods are a phylogenetically important group in the arthropod radiation for which relatively little developmental data are currently available. Our study provides valuable comparative data that complement the growing number of studies in insects, crustaceans and chelicerates, and is important for correct ancestral reconstruction and a fuller understanding of how and why germ cell development has evolved in different arthropod lineages.

C29-02 DNA methylation and phenotypic plasticity: Is DNA methylation a conserved mechanism for phenotypic plasticity in insects?

Duncan, Elizabeth (University of Otago, Dunedin, NZL); O'Neill, Meaghan (University of Otago, Dunedin, NZL); Dearden, Peter (University of Otago, Dunedin, NZL)

All animals respond to their environment, but some permanently alter their physiology, biochemistry and behaviour in response to an environmental cue; a phenomenon known as phenotypic plasticity. We have been using two insect models of reproductive phenotypic plasticity (the honeybee *Apis mellifera* and the pea aphid *Acyrthosiphon pisum*) to understand how plasticity is regulated and if there are common mechanisms that underpin these independently evolving examples of phenotypic plasticity. DNA methylation (the addition of a methyl group to a cytosine residue) is a key epigenetic modification associated with multiple molecular functions including regulation of alternative splicing of mRNA transcripts in many animals and regulation of gene expression in vertebrates. The mechanisms of DNA methylation are ancient and found throughout the animal kingdom, although the regions of DNA that are targeted in different species are diverse. DNA methylation may contribute to plasticity, particularly in highly fluctuating environments, and has been shown to be important for phenotypic plasticity in insects, i.e., caste development in the honeybee. The conservation of the DNA methylation system raises the possibility that DNA methylation may be a conserved mechanism underpinning phenotypic plasticity. Spatial and temporal expression of the DNA methylation and demethylation genes (*Dnmt1a, Dnmt1b, Dnmt3* and *Tet1*) is consistent with a role for DNA methylation in both these examples of phenotypic

plasticity. Further, functional manipulation of DNA methylation in the honeybee has shown that inhibiting DNA methylation enhances the transition from sterile to reproductive worker states, suggesting that demethylation or remodelling of the DNA landscape is required for this example of phenotypic plasticity. We are now working towards understanding how the methylation landscape is affected and changed in response to an environmental cue in both these species. Although the mechanism of DNA methylation is conserved, the landscape of methylation patterning is likely diverse with different genes targeted in the two species. Understanding how DNA methylation is functioning to regulate reproductive phenotypic plasticity in these species will give us insight into the evolution of phenotypic plasticity, providing evidence that either DNA methylation is an ancient and conserved mechanism of plasticity or it has been repeatedly co-opted over evolutionary time to mediate plasticity in different species.

C29-03 Modification of anterior-posterior patterning system toward fin-to-limb transformation: Origin of thumbs and radius

Onimaru, Koh (Tokyo Institute of Technology, Yokohama, JPN); Kuraku, Shigehiro (RIKEN Center for Developmental Biology, Kobe, JPN); Takagi, Wataru (University of Tokyo, Kashiwa, JPN); Hyodo, Susumu (University of Tokyo, Kashiwa, JPN); **Tanaka, Mikiko** (Tokyo Institute of Technology, Yokohama, JPN)

Pectoral fins of the catshark, *Scyliorhinus canicula*, which are composed of pro-, meso- and metapterygium, are comparable with those found in the majority of chondrichthyans, some basal actinopterygians, placoderms and acanthodians. According to anatomical studies, pro- and mesopterygium have been lost and the autopod (digits and wrists/ankles) emerged at the distal end of the metapterygium during the fin-to-limb transition. Here, we examine the fin development of catshark, and reveal that anterior-posterior patterning changes in fins would have triggered anatomical transformations such as digit acquisition and reduction of pro- and mesopterygium. Anterior radials of catshark fins expressed a gene set shared with the anterior elements of mouse limbs such as digit I, radius and deltoid process. Nevertheless, the expression domain of the anterior genes was more extensive in catshark fins than in tetrapod limb buds. Furthermore, homologous elements of *Gli3* anterior limb enhancer from both catshark and elephant shark Callorhinchus milli drove reporter expression throughout the limb buds of chick embryos. These results suggest that anterior-posterior patterning system may have been modified through alteration of the *cis*-element of *Gli3*, resulting in transformation of anterior fin radials into digit I, radius and deltoid process of limbs.

C29-04 Evolution of neurosecretory brain centres: A cell population with an apical-organ-like transcriptional profile pioneers the central nervous system in the centipede, *Strigamia maritima*

Hunnekuhl, Vera (Cambridge University, GBR); Akam, Michael (Cambridge University, GBR)

The apical plate of primary marine larvae is characterized by a common set of transcription factors comprising *six3, rx, hbn, nk2.1* and *FoxQ2*. It harbours the apical organ, a neural and ciliary structure with neurosecretory properties. A subset of the factors that specify the larval apical plate also characterizes the vertebrate forebrain including the developing neurosecretory hypothalamus, and a shared evolutionary origin of the neurosecretory cells has been suggested. It is under debate whether a territory homologous to the apical plate is present in the embryonic arthropod head. We identify an anterior medial tissue in the head the centipede that shares the transcriptional profile of the apical plate of marine larvae, including nested domains of *FoxQ2* and *six3* expression. This domain gives rise to a population of early differentiating neurons that erect a primary axonal scaffold of the central nervous system. These neurons do not belong to the *vsx*+ pars intercerebralis, but they express differentiation markers that characterize the apical organ of marine larvae including markers for neurosecretory activity. They also express markers of hypothalamic neurons, including *otp, vtn* and *vax1*, of which the latter is missing from insect genomes. We show that the centipede protocerebrum derives from two distinct developmental territories, an anterior medial and an ocular region. The cells at the anterior-most tip of the neuraxis, deriving from the anterior medial region, share characteristics of apical organ cells of invertebrates and with hypothalamic neurons of vertebrates, supporting an evolutionary relationship between all these structures.

17.20 – 17.35 Student Poster Prizes
ROOMS C1&2

17.35 – 18.15 Keynote Lecture (K4)

Ancient genes, mesoscale physics, and the origins of animal development

ROOMS C1&2

Stuart Newman
(New York Medical College, USA)
Chair: Frietson Galis

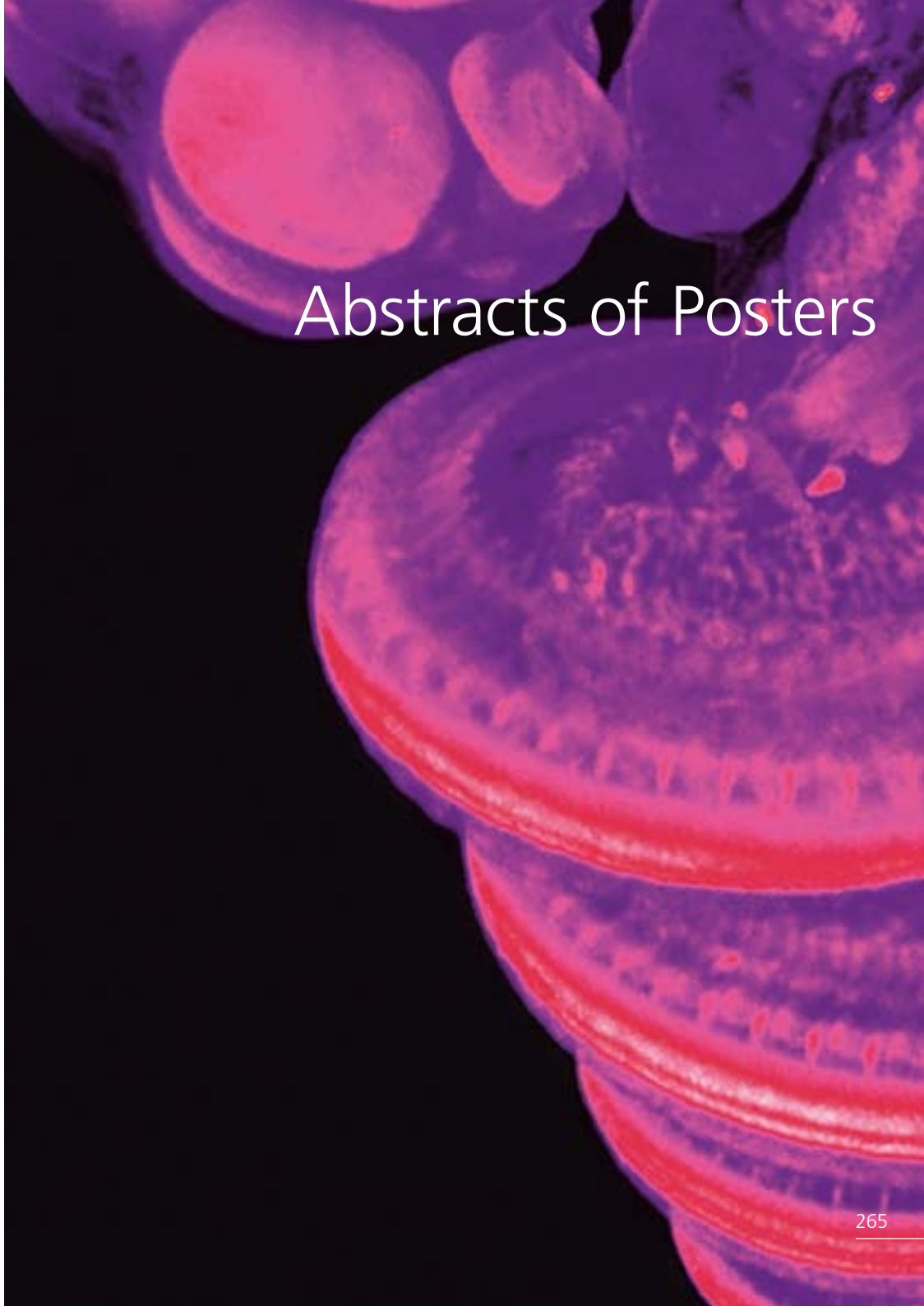
The metazoans, or multicellular animals, arose in several relatively rapid episodes, beginning in the Ediacaran period roughly 600 million years ago. The earliest stages involved the repurposing of the products of ancient genes of unicellular opisthonkts, organisms ancestral to both present-day fungi and animals. Some preexisting cell surface molecules (e.g., cadherins), owing to environmental change or mutation, came to mediate homophilic adhesive forces, producing multicellular aggregates. In the new multicellular context, other preexisting molecules (BMP, Hedgehog, Wnt, Notch, chitin, collagen, etc.) mobilized additional "generic" (i.e., common to living and nonliving systems) mesoscale physical effects (diffusion, lateral inhibition, differential adhesion of polarized subunits). The stereotypical morphological motifs (multlilayers, lumens, segments, appendages) generated by these "physico-genetic" processes provided developmental templates for the body plans and organ forms of all the subsequent animal phyla. The rise of the true metazoans, however, required the transformation of cell aggregates that were developmentally competent but genetically heterogeneous into genetic individuals. This occurred with the emergence of specialized, enlarged or matrix-secreting cells, proto-eggs, which were capable of generating multicellular clusters by cleavage or confinement, rather than aggregation, ensuring that the clusters were isogenomic and the organisms that developed from them evolutionarily more stable. The presence of an egg stage of development also enabled specification of the initial and boundary conditions of the physical effects mobilized during the multicellular "morphogenetic stage" (blastula, inner cell mass) of development, which made generation of phylotypic body plans increasingly reliable. Further evolution for robustness and reliability of development of organisms with established morphological phenotypes led to the emergence of "non-generic" physical regulatory mechanisms which, though preserving the generically originated forms, are unlike phenomena seen in nonliving matter. The perspective outlined provides plausible answers to several long-standing questions in evolutionary developmental biology: (i) Why did certain morphological motifs appear recurrently and independently over the course of animal evolution? (ii) Why are analogous structures often generated using the products of homologous genes? (iii) Why did morphological diversification of the animals occur in bursts, with little change in key genes of the developmental toolkit? (iv) Why are eggs of animals of different classes and genera within a given phylum often radically different in their morphologies and internal patterning processes? (v) Why do phylogenetically close and even morphologically indistinguishable species sometimes utilize widely divergent molecular and cellular pathways of development?

18.15 – 18.20 Conference Closing
ROOMS C1&2

18.20 – 19.10 EED Business Meeting
ROOM C1

19.30 Joint departure for Conference Dinner

Notes



Abstracts of Posters

Posters

P-001 A comparative genomics and transcriptomics approach to the study of *Hox3/zen* gene evolution in insects

Vargas Jentzsch, Iris (Cologne Biocenter, GER); **Gurska, Daniela** (Cologne Biocenter, GER); Panfilio, Kristen A. (Cologne Biocenter, GER)

Unlike canonical Hox genes, which function in embryonic tissue specification along the anterior-posterior body axis in all bilaterians, the insect orthologues, known as zerknüllt (zen), have undergone multiple instances of functional divergence as well as lineage specific duplication. All known zen genes are instead involved in the development of the extraembryonic membranes (EEMs). The EEMs have a protective role allowing embryos to survive in diverse ecological niches, hence contributing to the eminent evolutionary success of insects. Key evolutionary changes in EEMs correlate with changes in zen genes; it is only within winged insects that complete EEMs are observed as a morphological innovation, and that zen concomitantly acquired a strictly extraembryonic role.However, zen genes differ in the exact nature of their role, functioning either in early tissue specification or in late morphogenesis. The current distribution across the phylogeny of early and late functioning zen genes leaves open multiple hypotheses for how the observed evolutionary changes occurred. We focus on two well established species in our lab for grounding comparative analyses to elucidate these changes: the hemimetabolous bug *Oncopeltus fasciatus*, which has a single orthologue with a late role, and the holometabolous beetle *Tribolium castaneum* with two diverged paralogues, one early and one late. The *Oncopeltus* genome, and those of additional bug and beetle species, were recently sequenced and assembled in the context of the i5K genome project, and we use this genomic data to add important pieces to the zen phylogeny puzzle. Our bioinformatic work is complemented by experimental investigation of differential zen expression in early embryonic stages of *Tribolium*, which will be expanded to later stages and to *Oncopeltus* in future work. Moreover, subsequent knockdown (RNAi) and RNA-seq will reveal the degree of divergence in transcriptional targets between the paralogues and orthologues. This will help to draw a final image regarding whether the different roles of early and late zen genes arise upstream, downstream, or within the zen genes themselves. Ultimately, these diverse lines of evidence will help determine the ancestral role of zen within the insects, and to pinpoint when the subsequent functional changes and duplications occurred.

P-002 A comprehensive pipeline for identifiying lncRNAs on the basal-branching chordate Amphioxus

Herrera, Carlos (University of Barcelona, ESP); Rossell, Ariadna (University of Barcelona, ESP); Burguera, Demian (University of Barcelona, ESP); Irimia, Manuel (University of Barcelona, ESP); Garcia-Fernàndez, Jordi (University of Barcelona, ESP)

Among the numerous classes of RNAs, long non-coding RNAs (lncRNAs) are similar in terms of expression and gene structure to the mRNAs but lacking the potential to encode proteins. Over the last years, lncRNAs have been proven to play important roles in gene regulation, and shown to be involved in many key developmental processes. However, the evolutionary dynamics of lncRNAs has been scarcely studied, and few data are known at key evolutionary nodes of animal evolution, as the origin of chordates and vertebrates. The cephalocordate amphioxus is widely recognized as the best proxy for these nodes. Very little is known about the conservation of these developmental functions due to the low sequence conservation of lncRNAs, which has hindered the identification of deep orthologs among distantly related species. We aim to identify the lnRNA complement of the amphioxus genome, in order to approach the complement and evolution of this regulatory fraction of the genome at the origin of chordates and early evolution of vertebrates. For this, we used RNA-seq data from several adult tissues and developmental stages of *Branchiostoma lanceolatum* in order to identify its lncRNAs. First, we used CPAT software and blastx searches to eliminate transcripts with coding potential and/or with similarity to conserved protein domains upon 6-frame translation. Next, we selected only those with multiexonic gene structures (at least 3 exons) and a minimum length of 500 nucleotides. This pipeline yielded around 1500 putative lncRNAs and we will show the preliminary analyses of these Bl-lncRNAs, regarding their experimental validation, their conservation among amphioxus species and to other invertebrate and vertebrates, and their expression profile during development and in adult tissues.

P-003 A developmental model for variation in floral organ number

Kitazawa, Miho (Osaka University, Toyonaka, JPN); Fujimoto, Koichi (Osaka University, Toyonaka, JPN)

Morphological variations ubiquitously appear in continuous traits such as organ sizes and discrete numbers like body segments in multicellular organisms. The developmental sources of the variations,

however, are largely unknown. Floral organs such as sepals and petals exhibit a discrete variation even in an individual plant. The variation of floral organ number within a population of a single species tends to be asymmetric, namely, often increases but rarely decreases, or vice versa. We integrated field observations and statistical analysis to quantitatively show a developmental model for the asymmetric distribution. This model is derived from two hypotheses of floral development: Symmetric intraspecific variation of expression boundary of the homeotic genes that determine floral organ identity known as ABC genes, and a semi-concentric organ arrangement. Compared with several other models based on statistical model selection, the present model significantly fits the observed organ number variations from basal eudicots such as Ranunculaceae to core eudicots such as Caryophyllaceae, indicating that the asymmetric variation could be an indicator of the floral organ positioning and expression domain variation of homeotic genes.

P-004 A direct transgenic assay to identify highly conserved regulatory sequences acting in Drosophila development

Schmied, Christopher (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Kalinka, Alexander (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Tomancak, Pavel (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER)

Networks of regulatory genes, mainly transcription factors, organize the development of animals and shape their morphology. During the course of evolution the *cis*-regulatory elements of these genes and thus their expression are diverging or conserved. Previous work showed that during early and late embryogenesis of different Drosophila species, gene expression levels diverge. However, during mid embryogenesis, the arthropod phylotypic period, gene expression levels are conserved. This is the hourglass pattern of morphological evolution. This leads to the following question: Is this conservation also present in spatial and temporal patterns of gene expression? To address this, I will express regulatory genes from different Drosophila species in *Drosophila melanogaster* in trans and compare their expression patterns with the orthologous genes of *D. melanogaster*. I tagged transcription factors with C-terminal fluorescent tags using recombineering and insert them together with their endogenous locus into landing sites of *D. melanogaster*, using the φC31 integration system. I will image the embryogenesis of lines carrying the non-melanogaster gene, tagged with SGFP, and the melanogaster ortholog, labeled with mRFPruby, by SPIM microscopy. Finally, I will use 4D co-localization analysis to quantify the amount of conservation in the expression patterns.

P-005 A new self-regulatory model for DV patterning in a basal insect

Sachs, Lena (University of Cologne, GER); Chen, Yen-Ta (University of Cologne, GER); Roth, Siegfried (University of Cologne, GER)

In *Drosophila melanogaster* dorsal-ventral (DV) patterning requires Toll and BMP signaling to establish ventral and dorsal cell fates, respectively. While the function of BMP signaling is conserved throughout bilaterian animals, Toll signaling is believed to participate in DV patterning only in insects. To analyze the functions of Toll and BMP signaling components in the DV patterning system of a basal insect, we performed parental RNAi (pRNAi) in the milkweed bug *Oncopeltus fasciatus*. The expression of the ventral marker genes short gastrulation (sog) and twist (twi) was strongly reduced in Toll1 knockdown embryos, while it was expanded throughout the complete germ rudiment in decapentaplegic (dpp) knockdown embryos. However as we did pRNAi for Toll1 and dpp together the resulting phenotype resembled the one caused by the dpp single knockdown. These findings revealed that Toll signaling is necessary for DV polarity, but not for the activation of twi and sog, as it is in Drosophila. The BMP inhibitor sog is initially ubiquitously expressed. Its expression becomes only later successively cleared from the dorsal 65%, while it is enhanced in the ventral 35% of the germ rudiment. Therefore we propose that Toll signaling might only act as an enhancer for sog (and maybe also twi) expression on the ventral side and that the interplay between Sog and BMP signaling is mainly responsible for the patterning of the DV axis.

P-006 A 3D developmental atlas of *Euprymna scolopes* (Cephalopoda: Sepiolidae)

Klimpfinger, Claudia (University of Vienna, AUT); Metscher, Brian (University of Vienna, AUT)

The Hawaiian bobtail squid *Euprymna scolopes* is becoming one of the leading model organisms to study cephalopod development and evolution as well as animal-bacteria symbioses. Still there is only limited information available on its organogenesis. The existing papers deal with externally visible morphological features and major neural ganglia during development. To be a widely useful model organism a comprehensive map of organogenesis of *E. scolopes* is needed. This project is building a 3D image atlas of the developmental stages with annotations on organogenesis. The atlas will consist of high-resolution microCT images of contrast-stained specimens at developmental stages from early organogenesis to hatching. MicroCT images contain complete size-calibrated morphological information at resolutions comparable to low-power light microscopy (1-2 µm voxel sizes).

Because microCT imaging is non-destructive, the samples can be re-scanned with different staining or sectioned later for further detail. The volume images can be correlated with histological, ultrastructural, and gene expression data. The finished atlas, including detailed metadata, will be open access and can be expanded with further relevant images and information.

P-007 Adult prothorax patterning during insect typical metamorphosis

Hu, Yonggang (Georg August University of Göttingen, GER); Bucher, Gregor (Georg August University of Göttingen, GER)

Insects are the most species-rich taxon on Earth. Much of the holometabolous insect biodiversity is generated by changing body form during metamorphosis. During this process, the body shape experiences a dramatical change. Most insects re-use most of the larval epidermis to build the adult epidermis, while some structures are made by imaginal cells. *Drosophila melanogaster* is an extreme case: it only uses imaginal cells. In contrast, *Tribolium castaneum* shows more typical metamorphosis in that it re-uses larval epidermis for the adult structure. Thus it can become a model system to study the shaping of the adult body form. The genome-wide RNAi screen "iBeetle: Functional Genomics of Insect Development and Metamorphosis" is revealing genes required for pattern formation during metamorphosis. We have identified several genes that are required for forming the shape of the pronotum. Here, we collected 9 phenotypes affecting the pronotum from iBeetle database, 3 of which were confirmed by injecting non-overlapping dsRNA fragments into another beetle strain. The knocking down of Apaf-1 related killer gene in *T. castaneum* (Tc-Ark) results in the formation of a median line without bristles on the pronotum. The phenotype of gene Tc-001035 shows less sensory bristles. Furthermore, the lack of gene Tc-000401 induces anomalous indentations on the adult pronotum. In order to study these phenotypes in more details, we developed the proper in situ hybridization (ISH) approach for our research. We plan to combine the study of cellular process and gene function to elaborate the mechanism of these phenotypes.

P-008 An annelid homolog of the chordate notochord

Brunet, Thibaut (European Molecular Biology Laboratory, Heidelberg, GER); Lauri, Antonella (European Molecular Biology Laboratory, Heidelberg, GER); Handberg-Thorsager, Mette (Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, GER); Fischer, Antje H. L. (Harvard University, Cambridge, MA, USA); Simakov, Oleg (European Molecular Biology Laboratory, Heidelberg, GER); Steinmetz, Patrick R. H. (University of Vienna, AUT); Marlow, Heather (European Molecular Biology Laboratory, Heidelberg, GER); Tomer, Raju (Janelia Farm Research Campus, Ashburn, VA, USA); Keller, Philipp J. (Janelia Farm Research Campus, Ashburn, VA, USA); Arendt, Detlev (European Molecular Biology Laboratory, Heidelberg, GER)

The origin of chordates has been debated for more than a century. One key issue is the emergence of the notochord, a rod of rigid tissue that spans the dorsal midline of chordates. No homologous structure has so far been detected in any non-chordate bilaterian. Here, we identify the axochord, a ventromedian longitudinal muscle spanning the ventral midline of various non-chordate phyla, as a potential notochord homolog. Indeed, a primitively muscular nature of the notochord would be consistent with the fact that the amphioxus notochord is a striated muscle. We support this hypothesis by an in-depth molecular, developmental and morphological characterization of the axochord of the annelid *Platynereis dumerilii*. Just as the notochord, the axochord is positioned between the central nervous system and the axial blood vessel. It develops by convergence of bilateral mesodermal precursors that segregate from the neighbouring paraxial mesoderm through inhibition of the canonical Wnt pathway. We found that an identical, highly specific molecular coexpression cassette uniquely defines both the chordate notochord and the annelid axochord, including six transcription factors (brachyury, foxA, foxD, soxD, soxE, twist) as well as axon guidance signals (netrin and slit), extracellular matrix components (colA1, colA2 and laminin), and patterning signals (hedgehog and noggin). In larval worms, the axochord acts as an attachment band for lateral muscle blocks, enabling proper locomotion. We demonstrate moreover that an axochord can be safely inferred to have been present at each relevant node of the bilaterian evolutionary tree: within annelids, an axochord is virtually universally present, and molecular characterization of the sedentary annelid *Capitella teleta* demonstrates that its molecular profile is also conserved, just as the notochord profile is in chordates. Among other bilaterians, axochord-like ventromedial muscles have been described in the majority of bilaterian phyla, including for example molluscs, onychophorans, chaetognaths, and hemichordates. Altogether, the available evidence converges towards a ventromedial muscle being present in the urbilaterian ancestor, with a strikingly notochord-like molecular profile and morphology. We hypothesize that this structure became dorsal following dorsoventral inversion in chordate ancestors, and evolved into the notochord by vacuolation. We further suggest that loss of its muscular nature, further stiffening, and integration into a newly evolved rigid axial complex (the backbone), supported increased specialization towards undulatory swimming in early chordates and vertebrates.

P-009 Analysis of embryonic eye development in the spider *Parasteatoda tepiariorum*

Schomburg, Christoph (Georg August University of Göttingen, GER); Schacht, Magdalena (Georg August University of Göttingen, GER); Schneider, Julia (Georg August University of Göttingen, GER); Kirfel, Phillipp (Georg August University of Göttingen, GER); Posnien, Nico (Georg August University of Göttingen, GER); Prpic-Schäper, Nikola-Michael (Georg August University of Göttingen, GER)

The visual system is among the most important organs that facilitates the interaction of an organism with its environment. Spiders, like most arthropods, possess two independent visual systems, generally termed lateral and median eyes, respectively. These systems have a different developmental origin, are determined by different genes, show different morphology and function and, finally, have strikingly different evolutionary trajectories. We aim to reveal the embryonic origin of the median and lateral eyes respectively by analyzing the temporal and spatial expression of conserved eye development genes in relation to well characterized anterior head markers in the spider *Parasteatoda tepidariorum*. We show comprehensive expression data from the *P. tepidariorum* orthologs of the retinal determination network genes. Furthermore, we show differences in the developmental timing of the median and lateral eyes by analyzing the expression of Pt-rhodopsin1 as well as the head morphology of the developing embryo. The results are discussed with respect to the development and evolution of different eye types in insects and other arthropods.

P-010 Anatomical Network Analysis (AnNA) in morphological EvoDevo

Rasskin-Gutman, Diego (University of Valencia, Paterna, ESP); **Esteve-Altava, Borja** (University of Valencia, Paterna, ESP)

Morphological EvoDevo is a field of biological inquiry in which explicit relations between evolutionary patterns and the growth or morphogenetic processes are made. Historically, morphological EvoDevo results from the coming together of several traditions, notably Naturphilosophie, embryology, the study of heterochrony, and developmental constraints. A special feature binding different approaches to morphological EvoDevo is the use of formalisms and mathematical models. Here we will introduce anatomical network analysis, a new approach centered on connectivity patterns formed by anatomical parts, with its own concepts and tools specifically designed for the study of morphological EvoDevo questions. Riedl's concept of ''burden'' is tightly related to the use of anatomical networks, providing a nexus between the evolutionary patterns and the structural constraints that shape them. We describe our most recent advances in Anatomical Network Analysis of the tetrapod skull evolution and development, with a special emphasis on humans.

P-011 Axis determination and pattern formation in the pea aphid: Implications of the expression pattern of conserved developmental genes

Hsiao, Yi-min (National Taiwan University, Taipei, TWN); Chung, Chen-yo (National Taiwan University, Taipei, TWN); Lu, Hsiao-ling (National Taiwan University, Taipei, TWN); Cook, Charles E. (European Molecular Biology Laboratory — European Bioinformatics Institute, Cambridge , GBR); Chang, Chun-che (National Taiwan University, Taipei, TWN)

In the parthenogenetic and viviparous pea aphid *Acyrthosiphon pisum*, a temporal series of developing oocytes and embryos are enclosed within the ovarian tubules (ovarioles). Accordingly, developmental distribution of an mRNA during oogenesis and embryogenesis can be detected within an ovariole. In light of the developmental continuity from oogenesis to embryogenesis, we explored whether the anteroposterior (AP) and dorsoventral (DV) axes are preformed in the developing oocytes, as they are in Drosophila. Previously we identified anterior localization of hunchback (Aphb) mRNA in oocytes and early embryos of the asexual pea aphid, suggesting that the breaking of anterior asymmetry in the oocytes leads to the formation of the anterior axis in embryos. Restriction of the caudal (Apcad) mRNA in the posterior region occurs later during blastulation. However, when and where the DV symmetry breaks during development remains unknown. In this study we cloned and detected the developmental expression of decapentaplegic (Apdpp) and short gastrulation (Apsog), both of which play conserved roles for DV patterning in Drosophila and several other insect species. We expected to identify the asymmetric distribution of Apdpp and Apsog mRNAs, assuming they could implicate when and where the DV axis is established in the pea aphid. Asymmetric distribution of mRNAs encoded by the four Apdpp genes (Apdpp1-4) was not identified in oogenesis and early embryogenesis, whereas expression of Apsog could be identified in the ventral region of cellularized embryos. This suggests that Apdpp1-4 mRNAs do not play a role in dorsal patterning but Apsog mRNA is involved in ventral development in the pea aphid. In addition, we also detect the expressions of Hox complex genes labial (lab), proboscipedia (pb), Deformed (Dfd), Sex combs reduced (Scr), Antennapedia (Antp), Ultrabithorax (Ubx), abdominal-A (abdA), Abdominal-B (AbdB), expecting to understand the establishment of the body plan from axis determination to pattern formation in the pea aphid.

P-012 Axis patterning of the tick, *Boophilus microplus*

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Boophilus microplus is a cattle tick causing major economic losses to livestock production. Tick control using acaricides affects stages after hatching. Thus, strategies interfering with embryogenesis must be developed that depend on the knowledge of tick embryonic development. Most arthropod embryology has been investigated in the Diptera *Drosophila melanogaster* and the few molecular studies on chelicerates are restricted to spiders. Thus, investigation of the molecular mechanisms involved in tick oogenesis and embryogenesis is of great importance. In insects determination of dorso-ventral (DV) and antero-posterior (AP) axes starts during oogenesis involving a crosstalk between germ line and soma. This crosstalk is mediated by several pathways, including the EGF pathway. Whether this pathway also plays a role in AP and DV axis determination in *B. microplus* and by consequence in chelicerates is an open question. To answer this question, we used an antibody against MAPK (dpERK), a downstream effector of EGF. dpERK was observed in the oocyte nucleus region and in pedicels, suggesting that these latter cells act similarly to insects follicular cells. Several aspects of tick embryogenesis are similar to spider embryogenesis like cumulus cell formation and germ disc development, but many are different, like a transient ventral opening and fourth-leg regression in the end of development. At the moment an in situ hybridization protocol to identify the expression of major pathway components like dpp and its antagonist Sog (dorsoventral axis), Notch, Wnt8 (segmentation) is being developed. Functional analysis of these genes will be performed via RNA interference technique, which will provide important insights on the evolution of axis formation in arthropods.

P-013 Bio-cultural ontogenies in Mesoamerica: Data, epistemic issues, and the archaeological roots of EcoEvoDevo and niche construction theory

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A notion of bio-cultural phylogeny/phylogenetics seems to accomodate many recent discussions that address the links between eco-evo-devo, niche construction theory, and other theoretical constructs (e.g. gene-culture co-evolution) potentially useful to articulate the relationships between hominid/human cultures and 'biology'. As domains of empirical research, the origin of agriculture and the domestication of animal/plant species have provided suitable data to explore the extent of such theoretical connections (see, for instance, recent work by Laland & O'Brien, Piperno, and Smith). Using published information derived from decades of (mostly) archaeological research on plant domestication in Mesoamerica, here I evaluate if, and how, ideas growing from the aforementioned discussions do in fact improve our current understanding of bio-cultural phylogenetics in the area. As a potential contribution to the set of concepts already in place in the corresponding literature, I suggest that a notion of bio-cultural ontogeny might improve models of the seemingly stable configurations of multiple interacting species that characterize geographical areas where domesticates have originated during the Holocene. As a supplement, I briefly review the basics of the (mostly) archaeological theoretical frameworks that used to inform empirical work on plant domestication in Mesoamerica, in order to assess how far are eco-evo-devo and niche construction theorists going with respect to their predecessors, particularly in research areas connected to bio-cultural evolution.

P-014 Biophysical dynamic module for the polarization of auxin efflux carriers PIN-FORMED (PIN)

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How do physical and biochemical processes feedback each other during plant development? We try to addres this question by studying the polarization dynamics of auxin efflux carriers in the root apical meristem (RAM) of *Arabidopsis thaliana*. Auxins are important phytohormones that act as morphogens altering cell elongation/

proliferation, among other developmental processes. Auxin efflux transporters PIN-FORMED (PIN) are anisotropically located, i.e., polarized, in the plasma membrane and their patterns of spatial polarization correlate with the direction of auxin fluxes, thus, influencing establish the patterns of cell proliferation and elongation along the apical-basal axis of the root. Polarization of the PIN transporters is dynamic and can respond to environmental signals (e.g., light, gravity). Although some models try to explain PIN polarization dynamics, they are still unable to incorporate this dynamical richness. In this work we integrate experimental data for processes of different nature (e.g., chemical, molecular and mechanical) that interact among them in non-linear ways to polarize PIN transporters. Based on the reviewed information, we propose a dynamical module (sensu Newman and Bhat 2009) that takes into account feedbacks between biochemical and physical components such as ROP6 GTPase and the plasma membrane mechanical tension, respectively. The dynamical module has been modelled as a Boolean network to explore the role of the different components for the polarization of PIN efflux transporters. The model reproduces some essential features of PIN polarization dynamics and renders novel predictions, such as the relative role of the plasma membrane mechanical tension that we are presently testing in the laboratory. This work integrates uncoupled information and studies the overall regulatory logic behind the complex dynamic of PIN polarization. It also contributes to uncover generic mechanisms of plant morphogenesis focusing on the role of mechanical forces in these processes.

P-015 Brain development of the Hagfish, with reference to the vertebrate brain evolution

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The cyclostomes are composed of only two groups, lampreys and hagfishes. They form a monophyletic group, which diverged from gnathostomes (jawed vertebrates) over 500 million years ago. From an evolutionary perspective, lampreys are widely regarded as a valuable cyclostome model organism, because of the accessibility to their embryos. Due to the absence of some developmental compartments and regulatory genes' expression domains, embryonic brain of the lamprey has been regarded as representing an ancestral state of the vertebrate brain. Especially, the lack of Hedgehog and Nkx2.1 expression domain in the ventral telencephalon was thought to phenocopy mice mutants that lack medial ganglionic eminence (MGE). The MGE gives rise to GABAergic interneurons that migrate cortically and play important roles in ensuring the function of the gnathostome cerebral cortex. Also, the MGE itself develops into the pallidum, involved in the action selection. However, recent studies have suggested the presence of the pallidum and cortical interneurons in the telencephalon of the adult lamprey. Although there is a possibility that MGE develops after the embryonic development, it is very difficult to investigate their long-term larval and metamorphosis stage in the lamprey. On the other hand, little is known about the brain development of the hagfish, due to the difficulty in obtaining fertilized eggs. However, they have the advantage that they develop through direct development. In this study, we describe the brain development of the Japanese inshore hagfish, *Eptatretus burgeri*, and observed the expression patterns of vertebrate MGE markers. Surprisingly, in contrast to lampreys, hagfish Nkx2.1 and Hh genes are co-expressed in the ventral telencephalon suggesting the presence of MGE, as in gnathostomes. This suggests that the regionalization of the basal ganglia in the brain has been established before the divergence between cyclostomes and gnathostomes.

P-016 Can a subtle change in morphology propel groups of organisms into adaptive radiation?

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Transitions to new ecological habitats are often associated with adaptive radiation and species diversification. Acquisition of adaptive morphological traits can be a key element in the exploitation of new environments, and subsequent bursts of biodiversity. Water-walking insects, a group of hemipteran hemimetabolous insects that transited from terrestrial to water surface habitat, are a good example of species that experienced a burst of diversification after colonization of a new niche. Most diverse are the legs that allow them to exploit various niches of this new habitat, from small ponds to open ocean. However, the morphological and genetic changes at the origin of this ecological transition are unknown. We hypothesize that a subtle increase in leg length may have facilitated the transition to life on water by increasing the efficiency of the legs during water surface locomotion. To test this hypothesis, we aim to compare the efficiency of locomotion for different leg length phenotypes. First, we genetically manipulated leg length of *Microvelia americana* and *Mesovelia furcata* using RNA interference against the Ultrabithorax gene involved in regulating leg length. RNAi applied during nymphal development resulted in significant shortening of both mid-leg and rear-leg, without any apparent defect in the survival or behavior of the

emerged adults. Moreover these short-legged individuals are slower than control insects on both ground and water. Second, we compared different natural leg length phenotypes using *Microvelia* longipes species. Interestingly, individuals with longer legs are faster on water. These results demonstrate that traits that are key to animal ecological adaptation can be manipulated, using RNAi, and resulting individuals can be subjected to test of fitness. Moreover, longer legs increase the efficiency of locomotion on water and may be a key innovation at the origin of water-walking insect adaptation to life on water surface habitats.

P-017 Cell and tissue dynamics during *Tribolium castaneum* embryogenesis revealed by versatile fluorescence labelling approaches

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Studies on new arthropod models, like the beetle *Tribolium castaneum*, are shifting our knowledge of embryonic patterning and tissue morphogenesis beyond the Drosophila paradigm. In contrast to Drosophila, *Tribolium* embryos exhibit the short/intermediate germ-type, and they become enveloped by extensive extraembryonic epithelia, the amnion and serosa. The genetic basis of these processes has been the focus of active research. In our work, we have complemented genetic approaches with fluorescence live imaging of *Tribolium* embryos to make the link between gene function and morphogenetic cell behaviours during *Tribolium* blastoderm formation and differentiation, germband condensation and elongation, and extraembryonic development. I will first show that transient labelling methods result in strong, homogeneous and persistent expression of fluorescent markers in *Tribolium* embryos labeling the chromatin, membrane, cytoskeleton or combinations thereof. I will then demonstrate co-injection of fluorescent markers with dsRNA for live imaging of embryos with disrupted caudal gene function by RNA interference. Using these approaches, I will describe and compare cell and tissue dynamics in *Tribolium* embryos with wild type and altered fate maps. Our methodology provides a comprehensive framework to test quantitative models of fundamental patterning, growth and morphogenetic mechanisms in *Tribolium* and other arthropod species.

P-018 Cellular and molecular analysis of muscles in two Cnidarian species (*Nematostella vectensis*, *Aurelia aurita*)

Jahnel, Stefan (University of Vienna, AUT); Walzl, Manfred (University of Vienna, AUT); Technau, Ulrich (University of Vienna, AUT)

Muscles were a crucial innovation in the evolution of animals. In Bilateria muscles are a main derivative of the mesoderm. Cnidaria lack this intermediate germ layer, yet they possess muscle cells deriving from endoderm and ectoderm. Studying muscles in diploblastic animals might help to understand the evolution of mesoderm and muscle. In the last decade, the sea anemone *Nematostella vectensis* has been established as a new model system, however little is known about the histology and ultrastructure of the animal. So far it was unclear, whether muscles in *Nematostella* are still epithelial, or have become mesenchymal as in bilaterians. In order to understand the epithelial organisation of muscle cells we conducted electron microscopical studies. In addition we investigated the differentiation of muscle cells during early development of the animal. We found that the shape of the cells forming the muscle system varies considerably. Besides typical epitheliomuscular cells there are highly specialised retractor muscle cells, where the connection between the apical and basal portion is reduced to a long and thin cytoplasmatic bridge. Interestingly, tentacle retractor muscle cells detach completely from the surrounding epithelial cells and become mesenchymal basiepithelial cells attached to the mesoglea. Further we demonstrate that retractor muscles cell in mesenteries, as well as in tentacles differentiate from regular epithelial cells before they alter their epithelial organisation. In this context *Nematostella* gives novel insights into the question, how a transition from an epithelial muscle system to a mesenchymal could have happened.Recently, we have established methods for isolating muscle-cell specific RNA of *N. vectensis* and a scyphozoan jellyfish, *Aurelia aurita*. Initial results of the analysis of the muscle cell transcriptomes will be presented.

P-019 Characterizing the gene regulatory network active in early embryogenesis of the milkweed bug, *Oncopeltus fasciatus*

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Changes in Gene Regulatory Networks (GRNs) that control embryonic development may give rise to a wide variation of phenotypes in animals. Reconstructing these changes is therefore very important for understanding the evolution of animal body plans.

Within insects, the GRNs that govern Drosophila embryogenesis are well characterized, but if we are to study the evolution of GRNs and of the phenotypes that arise from them, we must adopt a comparative approach. Our research tries to elucidate the structure of the GRN governing early embryogenesis in *Oncopeltus*, focusing on the blastoderm stage. In Drosophila, the GRN that provides the initial patterning of the embryo is rather complex, with many known and defined mutual interactions and feedback. In *Oncopeltus*, the

picture is not as clear: the expression patterns of early developmental genes cannot be explained by the interactions between them that are known so far. This suggests that there are many components still to be discovered in this GRN. We have carried out a series of knockdowns of early patterning genes and followed the effects of the knockdown of one gene on the expression of other genes in the *Oncopeltus* blastoderm GRN. Among our results, we show that the genes orthodenticle and even-skipped play important roles in early embryogenesis: the knockdown of these genes leads to the most significant changes in the expression patterns of other genes. These results are different from what we know from Drosophila, where otd and eve have minor roles in initial patterning of the embryo. We are in the process of elucidating additional interaction, and hope to be able to ultimately reconstruct the complete blastoderm GRN in *Oncopeltus*, including distinguishing between direct and indirect interactions.

P-020 *Cis*-regulatory evolution and functional diversification of gene duplicates

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Gene duplication plays a major role in evolution of novel gene functions as it provides a material basis for variation and selection. We are interested in how *cis*-regulatory changes contribute to functional diversification ensuing gene duplication. To address this question we are studying the Ly6 gene family in insects. Members of this family encode different GPI-anchored membrane proteins and are fully conserved across drosophilids. Our analyses of the sequenced insect genomes indicate that a subset of these genes is unique to higher dipterans. We are focusing on seven paralogues in Drosophila, which we found to derive from sequential duplications of a single orthologue. By characterizing the embryonic tissue-specificities of the paralogues and their unduplicated orthologues in six insect species representing various stages of duplication, we determined how the original function of the unduplicated orthologue progressively diversified. Our results show that at each node of duplication, one paralogue inherited the tissue-specificities of the unduplicated orthologue, while the other paralogue acquired novel expression domains suggesting neofunctionalization. Moreover, we also found many instances of lineage-specific gains of tissue-specificities. We are currently identifying the *cis*-regulatory elements of some the duplicates and the unduplicated orthologues to elucidate the *cis*-regulatory mechanisms underlying the evolution of divergent expression patterns.

P-021 Commissureless regulation of Slit-Robo signalling in insects

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Slit-Robo signaling is a key mediator of axon guidance decisions in organisms ranging from planaria to vertebrates. Not surprisingly, Slit and Robo homologues can be identified in all insect genomes sequenced to date. In contrast to this conservation of ligand and receptor, organisms have evolved various mechanisms to regulate Slit-Robo signaling. In Drosophila, Commissureless is a key post-translational regulator of the Robo receptor that functions to prevent cell surface accumulation of Robo. Two additional Comm-family members are found in Drosophila and they vary in their ability to regulate Robo. We are investigating the evolution of Comm-like genes and regulation of Slit-Robo signaling in insects. Bioinformatic studies suggest that Comm-like genes are present in all sequenced Dipteran genomes, although the number of Comm-family members varies. Divergent Comm-like genes can be identified in representatives of Trichoptera, Coleoptera, Hymenoptera, Phthiraptera, Hemiptera, Blattaria, Ephemeroptera, and Odonata, but to date not outside of insects. The presence of a Comm-like gene in many insect orders suggests this gene was present early in insect evolution. There is evidence for three independent losses of this Comm-like gene: (1) the absence from all sequenced Lepidopteran genomes, (2) the absence in Tribolium but presence in more basal Coleopteran genomes, and (3) the presence in basal Hymenoptera, like the sawfly *Athalia rosae*, and absence in more derived Hymenoptera including ants and bees. In ongoing experiments, we are addressing the functional properties of divergent Comm-family members from a variety of insects using several approaches, including RNAi and a Drosophila cell culture assay for Robo regulation.

P-022 Comparative cephalochordate transcriptomics: Linking genotype and phenotype at the root of chordate evolution

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The basal chordate amphioxus is a key model system for understanding the evolutionary genesis and radiation of vertebrates. In this context, it has been long used in comparative anatomy and developmental biology. More recently, since the arrival of next generation sequencing,

it is also turning into an essential model system in comparative genomics. However, to fully exploit this opportunity, it is necessary to use these tools to assay the genome, transcriptome and epigenome of more cephalochordates. To this end, we have sequenced three different genuses of cephalochordates, complementing the ongoing sequencing of several species belonging only to the Branchiostoma genus. We have performed comparative analysis of RNA-seq and small RNA-seq data from *Branchiostoma lanceolatum*, Asymmetron and Epigonychtis. Using a combination of available and new computational analyses, we performed de novo assemblies and mapping of replicated datasets for each genus. The objective was to reliably reconstruct the molecular basis of mild but distinctive morphological differences between the three cephalochordate genuses. Our results suggest that such morphological differences, although mild, might have been critical for elaborating the definitive vertebrate body plan, as we find they are linked to differentially expressed transcripts between the three cephalochordates studied.

P-023 Comparative expression analyses of homeobox genes in mollusks

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Mollusca are the most speciose lophotrochozoan phylum and its representatives exhibit diverse body plans from creeping worm-shaped aplacophorans to highly mobile cephalopods with complex brains. Up to date only little is known concerning the developmental mechanisms that have led to the diversification of molluscan body plans during evolution. Homeobox genes such as parahox genes and pair-rule genes are involved in body plan patterning of many bilaterian clades. Previous studies on these genes have mainly focussed on ecdysozoan and vertebrate model species. Within the highly diverse Mollusca, however, only one gastropod and cephalopod species have been investigated. The present study focuses on the expression of the parahox gene gsx in the pygmy squid *Idiosepius notoides*, the scaphopod *Antalis entalis*, and the polyplacophoran *Acanthochitona crinitus*. A previous study showed that gsx is expressed around the gastropod mouth, in the apical organ, and in putative neuroectodermal cells. Besides taxon-specific expression domains in all three species, gsx is expressed in the cephalopod brain, the scaphopod and gastropod larval apical organ and probably neuroectodermal cells in scaphopods and polyplacophorans. In addition, expression of otx, pax2/5/8, and gbx was investigated in embryos of the cephalopod *I. notoide*s and pericalymma larvae and settled postmetamorphic specimens of the protobranch bivalve *Nucula tumidula*. In this talk homeobox gene expression in mollusks will be discussed with focus on the evolution of molluscan-specific organs.

P-024 Comparative transcriptomics to study habitat change and adaptive radiation in water striders

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Gerromorpha offer a good model to study the developmental genetic changes associated with the invasion of water surface habitat and their radiation into a diverse array of niches, from small ponds to open oceans. This ecological transition and subsequent specialization have been facilitated by substantial changes in morphology to meet the challenges and exploit the opportunities of the new environment. These morphological changes and specialization have captured a broad interest making Gerromorpha historically established models for ecology, evolution and biophysics. In particular, it is well established that the ability of this group to walk on water is mainly based on two traits: leg length and the hydrophobic bristles that cover the contact surface between the leg and water. However, despite the body of knowledge of the biogeography, phylogeny and biomechanic of water walking insects, the genetic changes underlying the invasion and radiations in water surface habitats are poorly understood. Here we generated and annotated the transcriptome of the water strider *Limnoporus dissortis* to study the genetic basis of leg morphology and specific adaptation associated with water surface locomotion. Using next generation RNA sequencing we identified the set of genes expressed in the legs during embryogenesis and tested for genes known to be involved in both bristle and leg development. Finally, after transcriptome data validation, we identified and analyzed a list of new genes potentially involved in second and third thoracic legs' length differences.

P-025 Composition of pre-nervous serotonergic signaling system in early embryonic development of sea urchin, clawed frog and mouse

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Neurotransmitters, such as serotonin, catecholamines and acetylcholine have numerous non-neuronal functions in addition to their classic one, and functionally active during early embryonic development,

long before the appearance of the nervous system. It is suggested that the primary function of these substances was humoral regulation of the functional state of the cell, and neurotransmitter function arose secondarily in nerve cells. Serotonin is commonly occurring in embryos at early stages of development. Pharmacological experiments on embryos of sea urchins have shown that serotonin is functionally active during the period of cleavage and is required for cell cycle regulation and blastomere interactions. Using molecular genetic techniques, we investigated the composition of the serotonergic system of early embryos three model objects belonging to different phylogenetic groups — the sea urchin *Paracentrotus lividus*, the clawed frog *Xenopus* and the mouse. Enzymes of serotonin synthesis are expressed at early stages of development, and it is the neural form of tryptophan hydroxylase that is presented in early embryos. Early embryos of all three species have a membrane transporter SERT performing the uptake of serotonin from the extracellular environment to the cytoplasm. Vesicular monoamine transporter VMAT is also expressed during early development of mice and frogs that is required for the accumulation of serotonin in the excretory vesicles and further intercellular signaling. It is interesting that in the early stages of development the Vmat2 gene is expressed, which is typical of the nervous system. Receptors are the key components of the serotonergic signaling system. In all species investigated several serotonin receptors were expressed simultaneously at early developmental stages. This may be associated with multifunctionality of serotonin at this stage of development. In the case of mice and frogs, receptors that are expressed on the early stages of development influence the same second messenger system (adenylate cyclase) in the opposite way. This fact may indicate a sensitive concentration-dependent serotonergic regulation of early development or its complex spatio-temporal organization. Our results suggest that the mechanisms of serotonergic signaling in early embryogenesis are generally similar to those in the nerve cells. However, the multiplicity of possible mechanisms of action is one of the characteristics of pre-nervous embryonic serotonergic system.

P-026 Conserved core and exchangeable modulators of the BMP signaling

Genikhovich, Grigory (University of Vienna, AUT)

BMP signaling plays a crucial role in the regulation of the dorso-ventral axis in triploblastic animals. In anthozoan Cnidaria, they also play a role in the specification and maintenance of the second, directive, body axis, which runs orthogonally to the main oral-aboral axis. We have functionally analyzed the molecular system of the directive axis maintenance in the sea anemone *Nematostella vectensis* by a combination of antisense morpholino-mediated knockdown, gene expression analysis and mathematical modeling. We show that the directive axis in *Nematostella* is maintained by two signaling centers, both expressing their own sets of BMP ligands and BMP antagonists. We demonstrate that a conserved core of the BMP signaling represented by BMP4, BMP5-8 and Chordin is responsible for the formation of the pSMAD1/5 gradient in the nuclei of the endodermal cells. The shape of the gradient is than modulated by other components of the system, such as Gremlin and Gdf5. Our model confirms the experimental observations that the core components of the system are very sensitive to change, while the modulators appear to allow for a lot of variation without collapsing the system. We speculate that this is the reason for the conservation of the core of the BMP signaling machinery and the exchangeability of the BMP signaling modulators in animal evolution.

P-027 Correlative, quantitative, and molecular 3D imaging for Evo-Devo

Metscher, Brian (University of Vienna, AUT)

Comparative studies of development depend on accurate visualization of morphology at size scales from whole embryos to ultrastructure, and microscopy methods for life sciences research have been driven by two opposing problems: the demand for ever finer spatial resolutions and the requirement to visualize tissues and structures in situ. At any given spatial scale, different imaging techniques have been optimized for either direct 3D imaging or for reconstructing 3D information from 2D images. In the size range most relevant for organism-based biological research (cm to sub-mm), lab-based x-ray microtomography (microCT) can produce 3D images of unsectioned tissues at histological resolutions for morphological, embryological, and even molecular studies. For animal embryos and other whole tissue samples, contrast-enhanced microCT imaging is a powerful complement to other 2D and 3D imaging methods, including light and fluorescence microscopy, light-sheet microscopy (SPIM), TEM, and SEM. Because tomographic imaging records quantitative spatial and object density information, it naturally generates data suitable for various kinds of modeling and analysis. Our current applications include measurements of developmental variation in zebrafish, correlating SPIM images of zebrafish cell proliferation with microCT images for development of asymmetric structure in the brain, and a new project to produce a 3D atlas of human embryos. We are also developing a new method for microCT imaging of molecular probes in whole-mount samples too large and opaque for fluorescence imaging.

P-028 CRISPR/Cas9 induced mutations in the Tribolium single-minded gene (Tc-sim) expose functions in ventral midline and growth zone patterning

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We found it difficult to generate knock-in mutations via homologous recombination by injecting cas9 mRNA and gRNAs into *Tribolium* embryos. As an alternative route to the generation of CRISPR/Cas9-induced mutations in the beetle germline that are linked to a selectable marker, we induced Tc-sim mutations via RNA injection into a GEKU strain that carries a 3xP3-EGFP insertion in a gene near the sim locus. By this means we obtained mutant strains that can be easily maintained and expanded for the efficient generation of mutant embryos. To this end, we designed 2 guide RNAs against Tc-sim using a simple algorithm (ZiFiT) and tested them for their mutagenic potential by inducing homozygous mutant clones in injected embryos. The gRNA that seemed more active in generating local sim-specific cuticle phenotypes was used for germline targeting. Despite substantial lethality rates among injected embryos, we obtained about 30 fertile G0 animals. From each of these we generated several offspring families that were inbred using the EGFP eye marker, and tested them for cuticle phenotypes ressembling the known embryonic RNAi phenotype of Tc-sim. About 20% or G0 animals gave rise to at least one sim mutant. The sim phenotype most prominently affects positioning of appendages and dorso-ventral polarity of the growth zone, an indication of a continued requirement for EGFR signaling during the germ rudiment extension process of this short germ insect.

P-029 Descriptive and statistical analysis of colour pattern evolution in two species of lizards

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Squamates (lizards and snakes) exhibit spectacular inter- and intra-specific colour variation generated by pigments (black/brown melanins, yellow and red pigments) and structural elements (guanine nanocrystals causing interference of light waves) incorporated into various types of chromatophores and iridophores, respectively (see e.g., Saenko et al. 2013). The adaptive value of colour variation in animals has been associated with thermoregulation, camouflage, predator avoidance, sexual selection, and speciation, although intra-specific polymorphism in colour traits can also involve pleiotropy, epistatic interactions and stochastic processes. Here, we investigate the shift in colour patterns occurring between juvenile and adult forms of two species of lizards (from the genera Eublepharis and Lacerta). For each individual investigated, we perform high-resolution 3D geometry and colour texture reconstructions at different time points during development from the juvenile to the adult stage. The different time points are non-rigidly aligned after semi-automated scale detection. The time evolution of colour patterns is then recapitulated on the 3D geometry. These analyses form the basis for characterisation and numerical simulation of pattern evolution in squamate reptiles.

P-030 Detecting homoplasy in the origin and evolution of adipose fins

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Adipose fins are appendages found in over six thousand species of ray-finned, or actinopterygian, fishes. These fins are positioned on the dorsal midline, posterior to the dorsal fin. Because of their apparently rudimentary anatomy, adipose fins are widely regarded as non-functional and possibly vestigial. Here, we test hypotheses of adipose fin origin, applying models of ancestral state reconstruction (maximum likelihood and maximum parsimony) to a recently published phylogeny of ray-finned fishes. These analyses find support for the hypothesis that adipose fins have evolved repeatedly, and this is consistent with the known distribution of adipose fins in the fossil record. As repeatedly evolved structures, we argue that adipose fins are likely adaptive, although their function is, as yet, unexplored. Additionally, we characterize adipose fin skeletons of 620 teleost species from 55 families to investigate skeletal evolution in the repeated instances of these new appendages. Adipose fin skeletons are highly homoplastic: anterior dermal spines, fin rays, and endoskeletal disks have all evolved repeatedly. Furthermore, patterns of skeletal evolution differ significantly between fin originations and challenge proposed models of skeletal evolution in new vertebrate fins. Adipose fins provide a promising new vehicle for evolutionary novelty, homoplasy, and principles of aquatic locomotion.

P-031 Development and evolution of asymmetric male genitalia in the *Drosophila nannoptera* species group

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How organs acquire their specific size and shape still remains largely unknown. It obviously involves tight regulation of growth and

proliferation, but how these ontogenetic changes actually occur and are integrated phylogenetically is not yet understood. We have identified a model system to examine the evolution of novel structures, which have evolved during the past 6-3 million years: the asymmetric male genitalia in the *Drosophila nannoptera* species group. In this evolutionary lineage, three out of four species possess left-right asymmetries in their male genitalia but each with distinctly affected organs or appendages. *Drosophila pachea* males exhibit an asymmetric pair of genitalia lobes, where the left lobe is 1.49 ± 0.08 (SD) times longer than the right lobe. By chance, we found a mutant with fully symmetric lobes and we use QTL-mapping to detect the mutation that causes the symmetric phenotype. Identifying the responsible gene will give an entry point to dissect the genetic basis of left-right asymmetry development. So far, we identified a genomic region in the *D. pachea* genome of several hundreds of kb that is associated with the symmetric lobe phenotype. We used a multi-locus dataset of gene coding regions of eight nuclear and three mitochondrial genes to estimate the phylogenetic relationships of the nannoptera group. We found that the four described nannoptera group species diverged rapidly, with very short internodes between divergence events. Our results indicate that a single evolutionary transition to asymmetric genitalia and to unusual sperm storage has probably occurred during evolution of the nannoptera group. Furthermore, our analysis of copulation behaviour shows that all tested nannoptera group species mate in a right-sided mating position whereas the outgroup *D. bromeliae* adopts a symmetric position. This indicates that a change in mating position occured ancestrally to the evolution of genitalia asymmetry in the nannoptera species group.

P-032 Development and evolution of dentition pattern for spatial order of tooth replacement in the Batoidea (Chondrichthyes)

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Although shark dentitions are well known for their generations of teeth and as isolated teeth in the fossil record, how these teeth are organized while building dentitions of enormous diversity is still poorly understood. This is especially true of the Batoidea (skates and rays) where due to rarity of embryos, developmental data is lacking. Hence, the aim is to compile data on tooth initiation, development and spatiotemporal order, from embryos to adults, from phylogenetically significant groups of batoids. Morphogenesis of the earliest development of the ray dentition is obtained from comparative developmental data of embryonic individuals of recent model organisms *Leucoraja erinacea* and *Raja clavata*. We used micro-CT to provide 3D volume rendered models (Drishti), with tissue segmentation in Avizo. Characters of the batoid dentition include presence of alternate addition of teeth as offset successional tooth files (families) versus single, non-alternate files in other chondrichthyans; presence of a symphyseal initiator region (symphyseal tooth present, versus two parasymphyseal teeth); a notable reduction to tooth addition along each jaw (proximally) restricting the total number of tooth families (labial to lingual tooth files), but retention of numerous successor teeth within each file. Similarities to the shark dentition (e.g., early presence of initiator symphysial region) indicate shared characters for the Elasmobranchii. For example, whether the alternate tooth addition dentition characteristic for the Batoidea is plesiomorphic or derived for Elasmobranchii, and Chondrichthyes as a whole, is still to be determined. These developmental morphological analyses will provide a solid basis to better understand dental evolution through diversity in these important vertebrate groups as well as the general plesiomorphic vertebrate dental condition.

P-033 Development of wing pattern diversity in *Heliconius butterflies*

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Heliconius wing colour patterns are one of the most diverse examples of phenotypic radiation and mimicry in nature. Recently, multiple genetic loci that control wing pattern phenotypes have been positionally cloned and associated with genes whose expression profiles appear to prefigure pattern. It is hypothesized that these pattern loci act as *cis*-regulatory elements, and are under selection to generate pattern diversity across the genus. Here, we seek to undertake experimental approaches that will allow the identification of the underlying molecular mechanisms of this pattern diversity, thereby examining the role of evolution of *cis*-regulatory elements in an ecological context.

P-034 Developmental basis of morphological diversity in the large African barbs from the Lake Tana (Ethiopia) species flock

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Lake Tana barbs species flock consists of 15 morphologically different species of g. Labeobarbus (Cyprinidae). This species flock is proposed to have a monophyletic origin from *L. intermedius* and to have

originated rather recently, about 15000 years ago. The most likely mechanism of fast divergence of Tana barbs is considered heterochrony —changes in developmental rate and timing. To verify this hypothesis, we investigated morphology and development of five barbs from the Lake Tana species flock : *L. intermedius, L. brevicephalus, L. megastoma, L. truttiformis*, and *L. crassibarbis*. By methods of geometric morphometry these species were shown to comprise four distinct groups differing significantly in the head shape: (1) *L. intermedius*; (2) *L. brevicephalus*, (3) *L. megastoma*, (4) *L. truttiformis* and *L. crassibarbis*. Comparison of cranial ontogeny has shown that in these species chondrocranium develops similarly, whereas bony skull ontogeny differs greatly in temporal parameters. Four patterns of bony skull development were revealed: (1) proposed ancestral one in *L. intermedius*; (2) accelerated in *L. brevicephalus*; (3) retarded in *L. megastoma*; and (4) accelerated in the early and retarded in the later ontogeny pattern of *L. truttiformis* and *L. crassibarbis*. Consequently, four distinct morphologies result from four patterns of ontogeny differing in temporal characteristics. Together, our data indicate participation of heterochronies in the morphological divergence of Tana barbs.

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P-035 Developmental pattern in mantis shrimps: Today and in the past

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Stomatopods, or mantis shrimps, are free-living predatory crustaceans inhabiting shallow marine or estuarine environment as well as the outer continental shelf or slope. Modern stomatopods (Verunipeltata) are characterised by their triflagellate antennules (i.e., first antennae), an articulated rostrum, highly efficient compound eyes and subchelate maxillipeds, with the second one developed as a massive raptorial claw. We currently know about 500 extant species. Stomatopod larvae possess one of the most aberrant sets of morphological traits among crustacean larvae. This includes numerous different structures, such as their large, fully functional raptorial maxilliped 2, their often large overall body size (up to 50mm in length), the elongated head region, or the hypertrophied shield. Generally, there are four different types of larvae recognized within stomatopods: Two early developmental forms, antizoea and pseudozoea, and two later developmental forms, the erichthus-type larva developing from either antizoea or pseudozoea, and the alima-type larva developing from squilloid pseudozoeae.

Despite the disparity of these larval forms both larval types develop into comparably similar-appearing adults. Hence, among modern stomatopods we find quite a disparity of developmental patterns. Here we highlight the morphologies of the two later larval types, the alima and the erichthus larvae. Certain subforms of the erichthus type appear to be ancestral (plesiomorphic) in many morphological aspects while the alima appears to be very derived. In addition to the comparison of the morphologies between these two modern larval types we also present the differences between modern morphotypes and the ones found in 150 million years old fossil deposits. For our approach we applied modern imaging techniques such as composite fluorescence imaging, making use of the autofluorescence capacities of the cuticle, to display also finest details of individuals of each of these groups. Based on these data we aim at reconstructing an evolutionary scenario for the emergence of larval specializations within Stomatopoda.

P-036 Developmental plasticity of germline development in the pea aphid *Acyrthosiphon pisum*

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The pea aphid *Acyrthosiphon pisum*, a hemimetabolous hemipteran insect with abundant adaptive capacity in response to specific environmental cues, has proven an excellent model for study developmental plasticity, and the published genome has made research on this species more accessible. Recent studies suggest that there are two distinct developmental programs directing axis patterning in the asexual viviparous and sexual oviparous embryos. In the germline cell lineage, we also found differential expression of duplicated piwi and ago3 genes occurred during the transition from asexual to sexual phases. This suggests that reprogramming of the Piwi-interacting RNA (piRNA) pathway, where Piwi and Ago3 proteins are involved, is required during the switch of reproductive cycles and that developmental plasticity may exist in the course of germline development. Using an affinity-purified antibody against the aphid germline marker protein ApVas1, nevertheless, we identified the preformed germ plasm in the uncellularized egg chambers of viviparous and oviparous embryos. This indicates that formation of germ cells in both types of embryos is germ plasm-dependent. In comparison with the piRNA pathway, germline specification does not display developmental plasticity in the pea aphid. However, whether components of the germ plasm differ in viviparous and oviparous embryos requires further investigation.

P-037 Developmental studies on the cucullaris muscle in the Mexican Axolotl (*Ambystoma mexicanum*)

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An important evolutionary feature of early tetrapods was the disconnection of the pectoral girdle from the head and the acquisition of a functional neck. This occurred just prior to the evolution of the first terrestrial vertebrates and therefore might be a precondition for the transition to terrestrality. Although the bony connection between the head and the shoulder girdle was reduced, a muscle connection via the cucullaris/trapezius remained, largely orchestrated by the neural crest (NC), as shown by data from mouse and chicken. The origin and character (head or trunk) of this muscle remains controversial in studies of amniotes. It has recently been shown, by using long-term fate mapping in the Mexican Axolotl (*Ambystoma mexicanum*), that the cucullaris is at least partly somitic in origin and that NC cells do not contribute to its connective tissue, which they do in amniotes. A main object of our research is to investigate the ontogenetic origin of this muscle further by testing whether the somatopleure contributes to the muscle, using classical extirpation experiments together with immuno-histo-chemistry and laser scanning microscopy for visualisation. Our first results show support for an, at least, partial lateral plate origin for the cucullaris muscle, and brings its trunk identity, suggested on the basis of fate mapping experiments, into question. Additionally, data on Pax3 protein distribution, give further support for a head origin of this neck muscle.

P-038 Differential canonical Wnt and Hh signaling in head and growth zone of a short germ embryo

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Canonical Wnt and Hedgehog (Hh) signaling pathways play a conserved role in posterior development of bilateria but their respective target gene sets have not been comprehensively identified so far. The long germ insect Drosophila does not show posterior elongation and early patterning occurs in the syncytial blastoderm where transcription factors diffuse between cells obviating the need for signaling pathways. In ancestral insect embryos, in contrast, patterning occurs in a cellularized environment where signaling pathways are predicted to play a more fundamental role and requirement of the Wnt pathway for posterior development has been well documented. Here, we use the short germ beetle *Tribolium castaneum* to investigate two putative Wnt and Hh signaling centers located in the anlagen of the anterior head and the growth zone. We find that Hh acts upstream of Wnt in the head, which is different from the Drosophila situation. In the growth zone, the interaction is opposite with canonical Wnt signaling acting upstream. We comprehensively determine the target gene sets of canonical Wnt and Hh pathways and distinguish the anterior from the posterior gene sets by genetically depleting head or growth zone, respectively. Surprisingly, there are significantly more targets in the growth zone than in the head for both pathways and we find that the respective target gene sets of the growth zone are essentially non-overlapping. We find that the Wnt pathway governs both patterning and growth zone metabolism by regulating genes required for epidermis, gut and mesoderm patterning including several pair rule genes, Tc-caudal, Tc-twist and hindgut patterning genes. Further, genes required for protein metabolism and cell division were regulated. Posterior Hh signaling activates several genes potentially involved in a proteinase cascade of unknown function. Unexpectedly, we find the Wnt target Tc-senseless to be required for hindgut development.

P-039 Disentangling oocyte polarity in panoistic ovaries. The role of Capicua in *Blattella germanica*

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Oogenesis in highly modified insects like *Drosophila melanogaster*, the classical model of meroistic ovaries, has been thoroughly studied. In contrast, the information available on the molecular mechanisms regulating oogenesis in panoistic ovaries (the ancestral ovarian type in insects) is notably scarce. In an attempt to afford data on the primitive ovarian type, we have been working on *Blattella germanica*, a hemimetabolous insect species with panoistic ovaries, in which only the basal oocyte is developed in each gonadotrophic cycle. We focused our research in those genes described as determinants of oocyte polarity in meroistic ovaries, and among them we studied the gene capicua (Cic), which encodes an HMG-Box transcription repressor factor that, in *D. melanogaster* ovary, is required for the establishment of dorsal-ventral polarity. The *B. germanica* Cic mRNA was identified and its expression pattern in nymphal and adult stages has been determined. Cic protein was localized in the egg chambers in both somatic and germinal cells. Depletion of Cic mRNA by RNAi determines changes in the anterior and posterior poles of the ovarian follicle and as a consequence, females become sterile. Moreover, data on the effects of Cic depletion on the expression of other genes involved in oocyte polarity was also revealed by the RNAi experiments.

P-040 Dissecting the genetic basis and evolution of form using haploid wasps

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Identifying and characterizing the molecular basis responsible for differences in form among species is one of the major goals of Evo-Devo. The Nasonia genus is an emerging model clade for such studies. Of the four species within the Nasonia genus, we are interested in *N. vitripennis* and *N. giraulti* for morphology studies. These species can be made interfertile in the lab by curing Wolbachia infections. *N. vitripennis* has a well sequenced and annotated genome. Additional sequencing is underway to make the genomic tools in *N. giraulti* comparable to those of *N. vitripennis*. *N. giraulti* is of particular interest as the head shape of the males is distinct from the other species. Further, Nasonia have haplodiploid genetics; fertilized eggs give rise to females and unfertilized eggs give rise to haploid males. This characteristic is particularly useful in the study of interspecies differences in male traits, since the haploid male progeny of hybrid females can be screened directly in the F2 generation. In addition to the interspecies shape differences, the haploid genetics of Nasonia is a particular advantage in mapping epistatic interactions among multiple loci. Hybrid crosses reveal negative epistatic phenotypes that are not present in either of the wild type species. We are testing the applicability of Multiplex Shotgun Genotyping, which should allow high resolution and high throughput genotyping of hybrid males. Lastly, we are characterizing the developmental basis of head shape in these wasps by examination and comparison of eye-antennal discs between species.

P-041 Dorsocross in the evolution of insect (extra)embryonic development

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Extraembryonic development is highly plastic across insect species. We investigated the role of one of its key regulators, the T-box transcription factor Dorsocross (Doc), in species possessing different modes of morphogenetic movements with respect to extraembryonic tissues. *Drosophila melanogaster* only possesses a single extraembryonic tissue (the amnioserosa) that covers the embryo dorsally and performs very limited morphogenetic movement. Here, doc is necessary for maintenance of the amnioserosa: knockdown/out results in a failure of germ band retraction. In addition doc has multiple roles in embryonic development including that of heart, wing discs and hindgut. In contrast, extraembryonic membranes (amnion and serosa) in the more basally branching holometabolous beetle *Tribolium castaneum* are highly dynamic, performing fusion, rupture and extensive movements. We found that knockdown of doc results in a disturbance of some of the key features of *Tribolium* extraembryonic development: impaired fusion (serosal window closure), ectopic rupture of membranes and failure of dorsal closure. However, the tissue intrinsic contraction seems not to be disturbed, albeit exerted ectopically due to failed fusion and rupture events. Furthermore, we could not find any disturbance of embryonic development or extraembryonic tissue specification.Embryogenesis in the even more basally branching hemimetabolous bug *Oncopeltus fasciatus* not only includes fusion, rupture and movement of the extraembryonic membranes, but also dramatic movement of the embryo itself. Here, rupture/withdrawal of extraembryonic membranes in late development (katatrepsis) seems to be impaired in doc knockdowns, resulting in a partial inside-out phenotype. Less severe phenotypes show disordered leg folding during katatrepsis, possibly linked to a distortion of the interplay between amnion and legs during preceding morphogenetic movements. Based on our comparison between Drosophila and more basally branching insect species, we propose that the ancestral role of dorsocross was in directing morphogenetic movements (including fusion and rupture) of extraembryonic membranes. We believe that additional functions linked to extraembryonic membranes (e.g., dorsal closure in *Tribolium*, amnioserosa maintenance in Drosophila) and independent of extraembryonic development (e.g., heart development in Drosophila) have been acquired in different species during insect evolution. Future research will reveal if our current view of the evolution of dorsocross applies across additional species. The transition of doc's function from extraembryonic morphogenesis to specification and maintenance of extraembryonic and embryonic tissues would be a fascinating example of an transcription factor acquiring new roles at earlier developmental stages.

P-042 Dual role of the canonical Wnt pathway in endoderm and posterior development in the brachiopod *Terebratalia transversa*

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The Wnt pathway is a master regulator of animal development and regeneration. While extensively studied in model organisms (e.g. vertebrates, Drosophila, *C. elegans*, planarians), little is known of the role of the Wnt pathway during embryogenesis in the vast majority of animal groups. This lack of knowledge hampers the reconstruction of the evolutionary history of the Wnt pathway in the Metazoa, and its role in major evolutionary events. Here, we characterise the

expression and function of the canonical Wnt pathway during the embryogenesis of the brachiopod Terebratalia transversa. Brachiopods (lampshells) belong to the Spiralia and are phylogenetically related to annelids, mollusks and planarians. During embryogenesis, they display radial cleavage, gastrulation by invagination and formation of a free-swimming larva that eventually settles and metamorphoses into the sessile adult. T. transversa has a stereotypical canonical Wnt pathway with 12 out of 13 of the Wnt ligands, 4 frizzled receptors, 3 out of 4 classes of Wnt inhibitors, and a full complement of intracellular components. These different elements are expressed throughout development along the animal-vegetal and antero-posterior axes of the embryo. Over-activation of the pathway with the GSK-3 inhibitor azakenpaullone from the 2-cell stage on induces the ectopic specification of endodermal/vegetal fates, a significant reduction of ectodermal/animal derivatives, and gastrulation failure. Treatment in blastula and early gastrula embryos, however, does not affect animal-vegetal patterning and gastrulation, but induces a severe embryonic posteriorization, reduction of anterior structures, and importantly, blocks the migration of the blastopore (opening of endomesoderm invagination) towards the animal pole to form the mouth, as it is observed in control embryos. Expression of the posterior markers evx and cdx in treated embryos indicate that the different blastoporal behavior is due to a posteriorization of mesodermal tissues, but not of the ectodermal cells defining the blastopore. Our results thus demonstrate a dual role of the canonical Wnt pathway in brachiopod development: an early function in endoderm specification, and a later function in antero-posterior patterning. Our data fills in the gap between the sparse knowledge of the function of the canonical Wnt pathway in the Spiralia (animal-vegetal specification in annelids and mollusks, but anterior-posterior regeneration in planarians), and noticeably, indicates that modifications in mesodermal patterning might be at the base of the diversity of blastoporal behaviors observed in the Bilateria.

P-043 Dynamic evolution of Crx-related homeobox loci in mammals: Birth and death from an unstable genomic region

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Developmental gene families are often very conserved and stable in animals, but bursts of family expansion, sometimes concomitant with major evolutionary transitions, are also apparent. Establishment of novel developmental gene families during early animal evolution could have proceeded by tandem gene duplication followed by "asymmetric" sequence evolution: after duplication functional constraints compel one of the two genes (the "parental gene") to retain ancestral features, by contrast, the other gene (the "daughter gene") diverges freely and rapidly until eventually becoming stabilized. However, much uncertainty remains concerning the mechanisms underlying this process. PRD homeobox genes experienced a striking increase in diversity in placental mammals, with a set of "novel" genes that have not been reported outside this clade. These include 4 new loci of uncertain evolutionary origin (DPRX, LEUTX, TPRX1 and TPRX2) located on the same human chromosome arm, 19q, and a related locus (ARGFX) on chromosome 3. We studied these loci in a wide range of species, comprising all major mammalian supergroups and using several experimental approaches (synteny analysis, coding and non-coding sequence conservation, phylogenetics and comparative transcriptomics). First, we uncover their evolutionary roots, demonstrating that the Otx-family gene Crx is the parental locus from which all are derived. Second, we show that most of the novel families exhibit a strikingly dynamic behavior, with frequent duplications, losses and limited sequence constraint, suggesting that their genomic environment is favorable to the birth and death of new duplicates. Third, we demonstrate that the functionally important stem cell and germ cell associated Obox and Crxos genes in rodents are in fact divergent members of the Tprx family. Finally, we propose a general link between gene family evolution and the architecture of their genomic location, and demonstrate how temporary loss of evolutionary constraints can contribute to the generation of sequence novelty and the origin of new genes.

P-044 Early leg development in *Tribolium castaneum*: The inside out phenotype of the new gene Tc-flipflop

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The general bauplan of the leg is a highly conserved structure among arthropods. Cytological and molecular processes involved in leg development are well studied in the fruit fly Drosophila melanogaster. The Drosophila leg originates from an imaginal disc that requires the initial invagination of cells. In contrast to Drosophila visible limb buds are formed in the red flour beetle Tribolium castaneum. This process involves the evagination of cells during embryogenesis similar to the leg development of vertebrates. Cell proliferation and cell shape changes contribute to the distal outgrowth of a leg. However, the molecular processes initiating the evagination of the limb bud are yet to be determined. To understand processes involved in early leg development we analyse candidate genes sharing similar mutant phenotypes such as inverted appendages in Tribolium. Our main focus lies on Tc-flipflop, a new gene identified during a RNAi-based screen.

Knock down of Tc-flipflop results in inverted legs forming inside the larval thorax rather than outgrowing distally. This can already be detected in embryonic stages forming the limb bud. Larval cuticles reveal fully developed legs including praetarsal claw. Tc-flipflop does not share any significant sequence homology to other species analysed so far. However, Tc-RhoGEF2 RNAi reveals a mutant phenotype similar to that of Tc-flipflop, indicating Rho associated signalling pathways to be involved in the early evaginating limb bud. We will further analyse the function of Tc-flipflop as well as other candidate genes on both cytological and molecular level in order to better understand the complex processes of leg formation.

P-045 Effects of artificially induced heterochronies on serial skeletal elements of teleosts

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Fish serial skeletal elements (SSE) (vertebrae and associated elements, scales, fin radials and rays, supraneural and infraorbital bones, gill arches and rakers, teeth, etc.) are widely used as features in the systematic and phylogenetic investigations. They display a high variety in number, shape, size, and functions within teleosts. One of the proposed evolutionary mechanisms of SSE diversification is heterochrony, changes in the developmental rate and timing leading to changes in the definitive morphology. To verify experimentally this hypothesis, the ontogeny rate of several fish species was artificially modified by the alterations of the level of thyroid hormones (TH), which via the control of target-gene expression influence the temporal characteristics of various ontogeny events. Changes in developmental rate and timing resulting from alterations in TH level were revealed to cause changes in the number and shape of most SSE in teleosts. The acceleration of the developmental rate leads to the decrease in SSE number, whereas the retardation causes the increase. Both acceleration and retardation change also the shape and size of SSE and, probably, may affect their functionality.

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P-046 Endoderm out of the head: Pharyngeal origin of cement glands and external gills in bichir

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Bichirs (Polypteriformes) represent a sister group of all extant actinopterygian fishes, however they share several important characteristics with sarcopterygians too. Here we describe thorough developmental and evolutionary analyses of most conspicuous bichir larval traits — cement organs and external gills. These structures represent transient but key adaptations that enable temporary attachment to a substrate after hatching, or facilitate larval respiration in less oxygenated water, respectively. Both develop early after neurulation as the first complex head organs, dominate a large part of head formation and alter standard schemes of early craniofacial development. Our analysis revealed the intriguing developmental potential of bichir head endoderm from which both these larval organs clearly derive. Cement glands originate as a pair of foregut diverticula by a process resembling pharyngeal pouch formation. These diverticula later detach as spherical pouches to finally open through outer ectoderm to secrete an adhesive mucus. This unique process of pouch detachment is likely apomorphic to this lineage. Similarly, external gills of bichir larvae were also found to develop on a basis of pharyngeal morphogenesis involving bilateral expansion of endodermal mass, which forms prominent external gill anlage. These however remain connected with foregut and covered with epidermis, making their origin distinct from cement gland. Whereas bichirs are commonly seen as an archaic lineage with variety of ancestral traits, our data show novel and strikingly apomorphic pharyngeal dynamics, which underlies the development of two distinct larval adaptations. As similar mode of foregut differentiation was earlier suggested to be present in other non-teleost fishes as well (bowfins, gars), our further efforts will be directed towards understanding the evolutionary history of this adaptive novelty among actinopterygians.

P-047 Evaluating if phenotypic classifiers capture genetic and geographical structure in Panther Chameleons (*F. pardalis*)

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Furcifer pardalis (the panther chameleon) is one of the most spectacular reptilian species in Madagascar and is found in a wide range of semi-humid and humid habitats along the northern and eastern costs of the island. One of the most striking features of the panther chameleon is its exceptionally large intra-specific colour variation: adult males have various combinations of bright red, green, blue and yellow, whereas females and juveniles are tan-brown with hints of pink or orange. This variation made *F. pardalis* a popular species on the pet market where colour "morphs" or "locales" are often named after Malagasy villages. Male coloration also varies with season and age, and local variation seems to exist within morphs. To test for a possible correlation between molecular phylogeography and male colour variation in panther chameleons, we preformed

the first and most extensive integrative analysis of phylogeography and phenotypic intra-specific diversity in *F. pardalis*. We sampled 324 individuals in Madagascar across the species distribution and used mitochondrial and nuclear DNA data to infer the genealogical relationships among populations. To test whether male colour pattern variation is correlated with population structure, we took high-resolution photographs of each sampled individual. The photographs were processed to calibrate for different lighting conditions and colour histograms in the HSV colour space were computed for predefined anatomical parts cropped from each picture. These colour histograms were then used to train a Support Vector Machine classifier and perform a hierarchical clustering analysis (based on Average Linkage Clustering algorithm) to investigate whether colour traits are good estimators of genetic relatedness and/or geographical location.

P-048 Evidence for an Inhibitory cascade in the development of limbs and digits

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The toe bones of most tetrapods include the metatarsal followed by a series of phalanges bones that act as a module in evolution and development. In the embryos, these bones develop in sequence as chondrogenic condensations that grow out distally and segment behind the growing tip to position the joints. By the time the tip is formed, the final adult proportions of the toes are achieved. Among taxa, phalanges' sizes covary in a highly predictable way, with variations ranging from equal-sized to a proximodistal gradient. Here, we have analyzed the variational patterns in size proportions of limbs and digits of a large number of vertebrate taxa that have up to 19 segments, in order to test general and specific predictions of the inhibitory cascade model, first described in teeth, in this skeletal system. We find support from both intraspecific and macroevolutionary patterns, as well as from experimental perturbations in the chick, for this type of developmental model.

P-049 Evo-diversification and evolution of integration: A phylogenetic approach to understand the evolutionary history of morphological traits

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Studying integration is essential to understand the evolution of shape, because the coherence of recognizable parts of most organisms is dependent on their developmental origin and structure. Drosophila wing morphology has been used extensively as an important model trait in evolutionary biology, since its genetics and development are well known. The present study tries to address questions about morphological integration of wing shape in an evolutionary context: (1) Does the Drosophila wing shape evolve across the Genus? (2) Does morphological integration evolve across the genus? (3) Is there any evolutionary integration in the Drosophila wing? Morphological changes were studied with geometric morphometrics to quantify shape variation and compare it between 59 species of Drosophila. Additionally, ordination analysis of Principal coordinates (PCoA) and comparative methods were applied to map shape data onto phylogeny. The results of the comparative analyses mapping the variation of covariance matrices onto the phylogeny showed a phylogenetic signal, which means that the pattern of integration evolves across the genus. There was also a clear evolutionary integration of wing shape. Thus, it was established that wing shape has strong internal covariation, and also that the integration process has evolved in the genus.

P-050 Evolution of bivalve by the modification of the cleavage pattern

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Bivalves evolved from their univalved ancestors of Mollusc and acquired two shell plates separated by ligament. Although bivalves develop through spiral cleavage patterns, the cleavage pattern of D lineage blastomeres is unique. This unique modification of spiral cleavage is directly associated with evolution of shell morphology, because they are thought to be derived from the descendants of 2d. To determine whether the unique cleavage pattern of bivalves is regulated depending on the interaction with other cells or by cell autonomous mechanisms, we performed cell isolation experiments and observed subsequent cleavage patterns of isolated blastomeres. When focusing on the largest derivatives of D blastomeres, 8% of isolated D blastomeres followed the cleavage pattern of normal development up to bilateral cleavage. We also examined the development of isolated blastomeres and found that isolated D blastomeres could develop shell plates, whereas larvae developed from AB blastomeres never had shell plates. Based on these observations, we concluded that D blastomeres control their unique cleavage pattern through intrinsic mechanisms and develop shell glands autonomously without any cell-cell interaction with other lineages.

P-051 Evolution of brachyury: Role in animal development

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Brachyury, a T-box transcription factor, is known to be a fundamental mesodermal gene in chordates. In vertebrates, as was recently

demonstrated by *in vivo* genome-wide regulatory analyses, Brachyury acts redundantly with other T-box factors and this complex not only promotes the activation of mesodermal genes in embryonic stem cells but also represses their potential neuronal fate. However, its ancestral role is found in the endoderm, more specifically as part of the gastrulation/invagination processes, but the exact molecular mechanisms that underlie this transition of Brachyury function in animal development remain unknown. To better understand the logics and evolution of the gene regulatory network (GRN) downstream of Bra in animal development, we used a genome-wide chromatin profiling (ChIP-seq) coupled with loss of function (RNA-seq) approach in two model organisms with a key phylogenetic position: the sea urchin *S. purpuratus* and the sea anemone *N. vectensis.* After raised and characterized anti-Brachyury specific antibodies for both species, we succeeded in establishing a Chromatin Immunoprecipitation protocol followed by high throughput genome sequencing. The revealed direct downstream targets of Brachyury were then cross-compared among the two species and the available data in the literature from vertebrates. This analysis allowed us to unravel conserved and divergent inputs of Brachyury with interesting evolutionary outcomes.

P-052 Evolution of cephalopod eyes by comparative transcriptome analysis of squid and nautilus

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Cephalopods are among the most advanced invertebrates, having sophisticated brains and eyes, dexterous chemo-sensitive arms, and exquisitely controlled dynamic coloration. Cephalopods are great models for understanding not only the evolution of these novelties but also as a unique and independent comparison to complex features in vertebrates. To understand the evolutionary process of the camera eye in squid and the pinhole eye in Nautilus, we have performed comparative transcriptome analysis of developing eyes of squid and Nautilus using next generation sequencer. First, although most upstream eye development controlling genes were expressed in both species, six3/6 that are required for lens formation in vertebrates was not expressed in Nautilus. Many downstream target genes of six3/6 were also not expressed in Nautilus, suggesting that deregulation of the six3/6 pathway led to the pinhole eye evolution in Nautilus. We also found five types of Pax-6 splicing variants but no duplication of the Pax-6 gene in squid. These variants show spatio-temporal patterns of gene expression during development. Previous studies have reported that vertebrate eyes are controlled by four Pax-6 splicing variants, whereas eyes are controlled by duplicated Pax-6 genes. Cephalopods acquired Pax-6 splicing variants independent of those in vertebrates and that these variants were similarly utilized in the development of the squid eye.

P-053 Evolution of early development of lophotrochozoa: Insight from lophotrochozoa specific homeobox genes

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Lophotrochozoa, including mollusk and annelid, is one of major group of bilaterian. Lophotrochozoa exhibits conservative and unique pattern of development known as "the spiral cleavage development". The character of spiral cleavage development is not only cleavage pattern with oblique angle. Segregation of developmental fate along A-V axis is also important and unique feature of spiral cleavage development. We searched molecular mechanism establishing these characters. We found lophotrochozoa specific new homeobox genes from their genome data. Most species have six or more genes in this family, which are probably gained by gene duplications. We examined expression patterns of these genes in mollusk (gastropod and bivalvia) and annelid (polychaeta). In these species, most genes exhibited a very transient zygotic expression in early cleavage stage (4-64cells stage). Most genes show localization in animal or vegetal blastomeres. Functional analysis indicated that these genes indeed perform essential role in segregating developmental fate along A-V axis. It is known that tandem dupulications of Hox genes were quite important for evolution of metazoa bodyplan. Likewise, we suppose that gain, duplications and diversification of lophotrochozoa specific homeobox genes were the key step for evolution of spiral cleavage development.

P-054 Evolution of germ line segregation and *Nanos* regulation in echinoderms

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Segregation of the germ line is a critical event in animal development, as it transmits heritable information to the next generation by creating sperm and eggs. This requirement could imply a constrained genetic program. However, evolution has produced a surprising diversity in mechanisms by which germ lines come to express a conserved suite of genes. These mechanisms can be loosely categorized within a continuum of maternal germ plasm inheritance vs. later zygotic induction modes. Echinoderms, a diverse and experimentally tractable phylum, provide a useful system for investigating this continuum.

We have performed expression and functional analysis of the germ line factor Nanos, and one of its targets, CNOT6, in a diverse set of echinoderm embryos. We infer that the ancestral mode of germ line segregation in echinoderms is by induction of a late-forming coelom, as observed in sea stars and sea cucumbers. However, the sea urchin uses early-forming vegetal cells, called small micromeres, which transcribe Nanos to enact a program allowing inheritance of maternal germ line factors. We now find that the micromeres of the pencil urchin, representative of the sister group to sea urchins, also accumulates Nanos, suggestive of germ line fate. We therefore infer that inherited specification of the germ line via micromeres is ancestral to the Echinoids. The question remains: what molecular changes drove the heterochronic shift of the germ line program into the micromeres? As in the embryos of many species, echinoderms display vegetally localized maternal Disheveled protein and activation of the canonical Wnt pathway. We are therefore testing whether the germ line module came under the control of this pathway, thus allowing for the shift into early-forming cells.

P-055 Evolution of metal response element (MRE)-binding transcription factors in three Branchiostoma species

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Adaptation to persistent trace metals depends on the induction of metallothionein (Mth) and metal transporters by MRE-binding transcription factors (MTFs). Mth induction in insects and vertebrates has been attributed primarily to MTF1, but recent evidence in oysters indicates a role for an MTF2-like protein as well. In order to clarify the potential mechanisms for metal tolerance in lancelets, we refined the annotation of candidate MTF1 and MTF2 genes and compared the patterns of codon usage in *B. belcheri, B. lanceolatum* and *B. floridae*. Phylogenetic analysis of both MTFs is consistent with the basal position of cephalochordates in the chordate lineage, and with the later divergence of a common ancestor of *B. lanceolatum* and *B. floridae* from the original lancelet ancestor. In all three species MT1s share the common domain architectures of 4 zinc fingers. MTF2, however, revealed a surprising organization in lancelet genomes. In *B. belcheri* the upstream zinc finger domains are separated from the downstream DNA-binding domain by a long intron encoding a GPCR-like protein on the reverse strand (a feature absent from the urochordate MTF2 gene). In *B. lanceolatum* and *B. floridae*, this GPCR-like insertion has evidently separated the upstream and downstream domains into separate genes, corresponding to separate cDNAs identified in the *B. lanceolatum* transcriptome. The patterns of synonymous codon usage in the terminal exon encoding the highly conserved, DNA-binding domain, were also found to be highly divergent: of 240 sites 10 sites were polymorphic in *B. floridae*, 8 in *B. lanceolatum* and 0 sites were polymorphic in *B. belcheri* (n = 13, 8, 8). These findings are consistent with a neofunctionalization of MTF2 unique to the cephalochordate lineage.

P-056 Evolution of pancreatic cell types: Insights from the sea urchin *Strongylocentrotus purpuratus*

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The pancreas is a vertebrate gland composed of two main tissues, the digestive enzyme-producing acinar cells and hormone-producing endocrine cells. The best characterized pancreatic hormone is the insulin, a molecule that belongs to the insulin superfamily, a group of evolutionary related proteins that includes also many insulin-like peptides (ILPs) in invertebrates. Little information is reported on ILPs and homologues of pancreatic endocrine and exocrine cell-types in non-chordate deuterostomes. In order to fill this critical gap in pancreas evolution, our aim is to analyze the orthologues of vertebrate pancreatic genes, both transcriptional factors and terminal differentiation genes, during embryo development in the sea urchin *Strongylocentrotus purpuratus*, which among the chordate sister group ambulacraria, has proven to be a powerful instrument for evolutionary studies. We identified three main homologues of pancreatic cell-types in the sea urchin embryo and larva. The spatial expression of ILPs found in the sea urchin genome (two paralogues named SpILP1 and SpILP2) and their predicted receptor (SpInsr) has been extensively studied. Remarkably, SpILP1 has been found to be expressed and localized in a group of cells of the larval stomach and intestine in a feeding-dependent fashion. Notably, the orthologous of Pdx1 and Cdx2/3, which are known to modulate insulin expression, are also localized in some of the ILP1 + cells and their expression if food-dependent. Furthermore, we characterized the spatial expression of the orthologues of pancreatic transcriptional factors (SpNgn, SpNeuroD, SpIsl, SpHnf1, SpPtf1a, SpMist1) and exocrine pancreas terminal differentiation genes (digestive enzymes such as SpCpa2L, SpPnlp, SpAmy3), which expression increases after feeding. Particularly, we found a peculiar 'acinar-like cell type' in the upper stomach of the sea urchin larva where both transcriptional factors and target genes are expressed. In addition, functional analysis showed that the link between Hnf1, Ptf1ta and the pancreatic digestive enzymes is conserved. Moreover, the already identified SpLox&SpBrn1/2/4+ cells have been further characterized and we found that it is a neurosecretory cell type that produces a novel neuropeptide. In

conclusion, comparing the above outcome together with available data in other animal models, we propose a model of pancreatic cell types evolution across deuterostomes.

P-057 Evolution of placode-derived neurons assessed by cell type-specific transcriptional profiling

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A major challenge in understanding the evolution of the vertebrate body plan is to model how gene usage evolved to produce the cranial placodes from which the paired sensory organs arise. Although our knowledge of placode development is growing, the function and evolution of the genetic regulatory networks underpinning the specification of differentiated cell types are not well known. In particular, we have been lacking specific molecular markers for the placode-derived neurons. Combining dissection, FACS sorting and next-generation sequencing in chicken, we have established cell type-specific transcription profiles in order to study the evolution and development of sensory neuronal cell types. Expression data is correlated to evolution of gene families by mining of several vertebrate and invertebrate genomes.

P-058 Evolution of Rab32/38 subfamily and their role in pigment cell formation in Chordates

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In vertebrates, Rab small GTPases have a crucial role in the regulation of vesicular transport and several members of the family are implicated directly in melanosomal physiology, including biogenesis and transport of melanosomes. Rab32 and Rab38 in mouse, localize mostly to secretory vesicles of the trans-Golgi network where they function redundantly to transport vesicles containing melanogenic enzymes, during melanosomes formation. The aim of our study is to investigate the evolution of Rab32 and Rab38 in Deuterostome evolution and their role in two aquatic chordate model systems: the cephalochordate Branchiostoma lanceolatum (amphioxus) and the teleost fish *Danio rerio* (zebrafish). Analysis of genomic data allowed us to identify two Rab32/38 orthologs in amphioxus and five in zebrafish (clearly two Rab32 and three Rab38) originated from different duplication events. The comparison of protein sequences among Deuterostomes, corroborated from phylogenetic analysis, showed new insights into Rab32/38 origin and especially clarified their identity within the subfamily. Moreover, on top of known Rab domains we have identified a novel ultra-conserved stretch of sequence, at the 3' of the Switch I domain, resulted exclusive of Rab32/38 subfamily, which we named FALK and whose function is still unknown. We are studying the role of two amphioxus genes during embryogenesis, focusing in particular on the comprehension of pigmentation structures development as the pigmented eye and the Hesse structures along the neural tube. We discovered a diversified expression pattern for these genes during zebrafish development, in particular they resulted expressed in epidermal pigmented cells (melanophores, iridophores, xanthophores) and in the retinal pigmented epithelium (RPE), that are the pigmented structures present in zebrafish embryos. It has been suggested that in zebrafish the extra duplicated genes, deriving from the fish specific whole genome duplication (FSGD) are probably subfunctionalized. Our findings of expression profiles of the different Rab32 and Rab38 suggest that the gene duplication in this family could have resulted in a gain of complexity in the pigmentation pattern and could correlate with the tremendous diversification of the skin coloration patterns in fishes.

P-059 Evolution of sex determining systems in the genus Silene

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The plant genus Silene (namely section Melandrium) has become a model for evolutionary studies of sex chromosomes and sex-determining mechanisms in plants. In spite of this, the subsection Otites that includes even more dioecious species then the section Melandrium remained neglected for a long time. Studies in *S. colpophyllia* and *S. otites* show that dioecy and the sex chromosomes in these species (subsection Otites) evolved independently from those in *S. latifolia* (section Melandrium), and we have further found that the sex-determining system in *S. otites*, a species closely related to *S. colpophyllia*, is based on female heterogamety, a sex determination system that is unique among the Silene species studied to date. Our phylogenetic data suggest that a switch from XX/XY sex determination to a ZZ/ZW system (or vice versa) probably must have occurred in

the subsection Otites. This is the first report on two different types of heterogamety within a mostly non-dioecious plant genus. Current availability of high throughput sequencing technologies enables us to study evolution of these sex determining systems in detail.

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P-060 Evolution of the GRN underlying eye differentiation in closely related Drosophila species

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A major question in evo-devo research is how tightly controlled gene regulatory networks (GRNs) can vary across related species to produce different morphologies while retaining functionality. One fascinating example is the insect eye, which shows a great variation in size and shape across the whole phyla. The compound eye of Drosophila has been thoroughly studied and some key players of the GRN that govern its development are known. However, the wiring of this network is not fully understood, and especially how this regulation varies between species. In our study we use three closely related species of Drosophila that show significant differences in eye size: *D. mauritiana, D. simulans* and *D. melanogaster*. We apply RNA sequencing on eye-antennal imaginal discs of different relevant larval stages for the abovementioned species. The use of next-generation sequencing for interspecies analysis poses also technical challenges. We have developed a method based on reciprocal annotation to be able to interrogate the full transcriptome and avoid species bias. We use this to analyze conserved genome wide changes in gene expression between stages in order to unravel those genes and molecular pathways involved in eye development. At the same time, by comparing gene expression among species we can detect those genes that are responsible for the differences in eye size. Additionally, by sequencing viable F1 hybrids between these species we also shed light on the regulatory mechanisms that create the divergence in gene expression. Surprisingly, our first results show that in early stages most of the expression differences in these closely related species are due to variation in trans, rather than cis. This complex dataset will be comprehensively combined in order to unravel the evolving nodes of the GRN.

P-061 Evolution of the molecular composition of the Nasonia dorsal-ventral patterning GRN

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The Gene Regulatory Network (GRN) responsible for Dorsal-Ventral (DV) patterning in *Drosophila melanogaster* is a well-studied model for how nucleic acids and proteins can coordinate to form a complex pattern. Since this GRN is so well known, it is logical to examine equivalent GRNs in other species to see how they have changed in the course of evolution relative to *Drosophila*. *Nasonia vitripennis* is a rising model organism whose genome has been sequenced, and is amenable to many molecular and genetic tools. A comparison of gene expression patterns has shown that while cell fates are nearly identical just prior to gastrulation, they diverge both up (pattern formation) and downstream (morphogenesis) of that stage. For example the relative importance of Toll and BMP signaling is inverted in *Nasonia*, with BMP playing the dominant role. Analysis of the factors downstream of these pathways uncovered further divergence between the DV GRNs of fly and wasp. Preliminary analysis has shown that some of the important players of the *Drosophila* GRN are conserved, some have new distributions, and some are absent in *Nasonia*. In addition, the *Nasonia* GRN has novel components that are not used for DV patterning in *Drosophila*, or are absent in the fly genome. The goal of this study is to utilize parental/embryonic RNAi and CRISPRs, combined with fusion proteins, in situ hybridization, immunohistochemistry, and live image confocal microscopy to fully characterize the *Nasonia* DV GRN. Focus will be on the role of the Dorsal Protein, the mechanism of a Toll independent feed forward loop leading to the expansion of ventral targets of Dorsal, and the roles of horizontally transferred endosymbiont genes that have been integrated into the *Nasonia* DV GRN.

P-062 Evolutionary conservation of leftward fluid-flow in left-right axis formation

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The establishment of left-right asymmetry from a bilaterally symmetrical early embryo depends on a leftward acting extracellular fluid flow, as shown in most vertebrate species examined so far. This flow is generated by a left-right organizer, a transient monociliated epithelium located at the posterior end of the neurula stage embryo, close to the embryonic organizer, Hensen's Node. We and others could show that directly downstream of this event, the Cerberus-like inhibitor of the TGF-beta growth factor Nodal, Dand5, is post-transcriptionally down-regulated. In turn, this results in the activation of Nodal signaling only on the left side of the embryo. The unilateral Nodal activity — which mediates asymmetric visceral organogenesis — has not only been demonstrated in all vertebrate species examined so far, but is also present in other Deuterostome clades like Tunicata, Cephalochordata and Echinodermata, and recently also in snails, i.e. a Lophotrochozoan species. Thus, we like to suggest the probability that a vertebrate-like

leftward fluid flow mechanism also acts as the symmetry-breaking event in other Deuterostome lineages. This is especially supported for the Cephalochordate amphioxus by the recent finding of a left-sided downregulation of a Cerberus-like ortholog.

P-063 Evolutionary novelty, a concept still in search of a definition

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The definition of the concept of evolutionary novelty poses several ontological and epistemological problems. A commonly accepted intuitive definition of novelty exists: an evolutionary novelty is a phenotypic trait bringing a qualitative difference, rather than a quantitative one, compared to traits already present in the lineage. This definition can serve as a starting point but is insufficient because of the several possible ways to interpret the notion of qualitative difference. Some authors have insisted on the need to produce a definition of novelty as theory-independent as possible (e.g. Müller & Wagner 1991), in particular neutral towards the mechanisms responsible of the origin of novelties. However most definitions have to rely on theoretical terms to cash out the quantitative/qualitative distinction, such as function (e.g., Mayr 1960; Pigliucci 2008) or homology (e.g., Müller & Wagner 1991; Müller 2010). The distinction between the description of novelties and their explanation is often waved as a methodological principle, but its very possibility and its epistemic value is rarely overtly discussed. A good example is the influential definition of novelty as a "structure that is neither homologous to any structure in the ancestral species nor homonomous to any other structure of the same organism" (Müller & Wagner 1991). This definition is at odds with the dominant definition of homology in systematics according to which each trait can in principle be homologized (e.g., Wiley & Lierberman, 2011). To restrict the extension of the concept of homology, Müller and Wagner rely on a "biological concept of homology" (Wagner 1989) that is grounded in a theory of how trait develop and that, in consequence, is not neutral towards the mechanisms of the origin of novelty. Even if a restrictive view of homology is adopted, the establishment of traits with no homolog faces epistemological problems with ontological consequences? The grain problem (Cracraft 1990) applies to the taxonomic level at which novelties are individuated? Many examples of novelty are defined at high taxonomic levels when only the species level would be adequate. The asymmetry problem is related to the evidence on which claims of absence of homology are based? Evidence of the existence of intermediate forms falsifies a hypothesis of evolutionary novelty while the absence of evidence of intermediates does the not falsify a hypothesis of absence of novelty. Another strategy is to define novelty or, more precisely, different types of novelty by the specific mechanisms responsible for their origin (e.g., Müller 2010). Epigenetic mechanisms, phenotypic plasticity or the overcoming of strong developmental constraints are candidates. But because they can have clearly quantitative effects as well as apparently big qualitative effects, these mechanisms also prove insufficient. It is concluded that this absence of a satisfying definition does not threaten novelty as a general explanandum of life, but threatens evolutionary novelties as a real kind of evolutionary events.

P-064 Evolutionary origin and diversification of epidermal barrier proteins in amniotes

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Terrestrial vertebrates require an efficient barrier against trans-cutaneous water loss. In mammals, this barrier is built up by epidermal keratinocytes that express unique structural proteins encoded by a gene cluster, the epidermal differentiation complex (EDC). To get insights into the evolution of the skin barrier, we screened for homologs of the EDC in non-mammalian vertebrates. By comparative genomics, de novo gene identification and gene expression analyses, we show that, in contrast to fish and amphibians, reptiles and birds have an EDC comprising genes that are specifically expressed in the epidermis and in skin appendages. The relative arrangement, the peculiar exon-intron structures of EDC genes and the distribution of conserved sequence elements suggest that EDC genes originated by gene fusions and subsequently underwent highly divergent sequence evolution. An important component of the mammalian skin barrier, i.e., loricrin, was also detected in reptiles and birds, indicating an origin in a common ancestor of modern amniotes perhaps during the acquisition of a fully terrestrial lifestyle. Moreover, we identify genes that are specifically expressed in embryonic skin and in feathers, suggesting a new evolutionary-developmental scenario for feathers. Taken together, our results point out the important role of the EDC in the evolution of the epidermal barrier and of the skin appendages in amniotes.

P-065 Evolution-development congruence in pattern formation dynamics: Bifurcations in gene expressions and regulation of networks structures

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Deciphering and understanding potential relationships between evolution and development has been an important research goal for

over a century. Recently, dynamical-systems analysis has proven to be relevant to both development and evolution, and it may therefore provide a link between the two. Using extensive simulations to evolve gene regulation networks that shape morphogenesis, we observed remarkable congruence between development and evolution: Both consisted of the same successive epochs to shape stripes, and good agreement was observed for the ordering as well as the topology of branching of stripes between the two. This congruence is explained by the agreement of bifurcations in dynamical systems theory between evolution and development, where slowly varying gene-expression levels work as emergent control parameters. In terms of the gene regulation networks, this congruence is understood as the successive addition of downstream modules, either as feedforward or feedback, while the upstream feedforward network shapes the boundary condition for the downstream dynamics, based on the maternal morphogen gradient. Acquisition of a novel developmental mode was due to mutational change in the upstream network to alter the boundary condition.

P-066 Evolving modular gene networks in a multicellular context

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The modular structure of gene networks enables the re-use and combination of existing functionality, in some cases permitting the rapid evolution of phenotypic change with few mutations. An important question, then, is how this modularity evolves. A number of very general hypotheses about the evolution of modularity in gene networks have recently been demonstrated using simple models. Here, I outline a novel method for evolving modularity in gene networks that specifically mimics the demands of multicellular development. Populations of gene networks are selected to respond across a range of environments to simultaneously (1) integrate multiple cues to establish what environment they are in, and (2) coordinate a pattern of gene expression on the basis of this environment. This selective regime proves sufficient to separate the network into two modules: an upstream module that integrates the environmental cues to calculate position, and a downstream module that coordinates the correct gene expression on the basis of this position. A single regulatory product typically provides an „interface" between the two modules, compressing the positional information from the environment, and initiating a cascade of downstream gene expression. I show how more complex examples of this basic selective regime permit modular re-use, where positional information is combined using the boolean logic of *cis*-regulatory modules, and wired to existing downstream functionality. These results resemble recent empirical work on pigmentation gain and loss in fruit flies; work that has formed the basis of speculative „principles" of regulatory evolution. This modeling work thus provides in-principle proof that we can study fundamental problems of the organization of gene regulatory networks *in silico* in way that integrates with empirical work, and allows us to derive testable predictions that can be experimentally verified.

P-067 Exploring developmental cranial integration in great apes

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Extant great apes share a developmental pattern of endocranial shape change following the eruption of the deciduous dentition. As the endocranium arises as the result of a complex interplay between bone, meninx and the expanding brain, its development is the result of an integrated process. Exploring whether a shared pattern of cranial integration underlies this observed pattern of endocranial development would contribute to our understanding of the evolution of the developmental interrelationships between the brain and the cranium. Furthermore, by investigating modifications to a putative shared pattern of developmental integration we can better comprehend the ontogenetic mechanisms enabling the evolution of novel adult cranial morphologies. Here, we hypothesize that the shared pattern of endocranial development in extant great apes is driven by integration between the endocranium and the face via the cranial base. We segmented computed tomographic scans of dried crania to generate virtual endocasts and captured shape information of the face and outer neurocranium (574 landmarks and semilandmarks) as well as the endocranium (309 landmarks and semilandmarks). Thereby, we characterized the interspecific patterns and degrees of cranial developmental and evolutionary integration in ontogenetic cross-sectional samples of *Pan* (n=56), *Gorilla* (n=65) and *Pongo* (n=72). Following Procrustes superimposition, two- and multi-block partial least squares analyses were used to quantify the patterns and degrees of integration. Our partial least squares results indicate that while variation exists in the degree of integration between taxa and across ontogeny, extant great apes share a pattern of ontogenetic covariation. This shared great ape pattern likely reflects an ancestral integration pattern and thus provides a model for estimating cranial covariation in the primate fossil record.

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P-068 Exploring floral patterning in Arabidopsis with dynamic models

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The floral meristem in *Arabidopsis thaliana* is a determinate structures that produces four kinds of organs: sepals, petals, carpels and stamens. In *Arabidopsis,* floral identity is established by the expression of the master regulatory gene LEAFY, which triggers a genetic cascade leading to proper floral morphogenesis, including floral organ identity. Organ identity is determined by the ABC model, wherein the expression of the A-, B- and C-class genes in partially overlapping concentric rings yields four domains with distinct combinations of genes. The A-class genes APETALA1 (AP1) and AP2 are expressed in the two outer whorls and determine sepal and petal identities, whereas the C-class gene AGAMOUS (AG) is expressed in the two innermost whorls and determines carpel identity. The ABC model states that the A- and C-class genes have a mutually inhibitory effect, and that this mutual inhibition is crucial to the establishment of their final expression domains. We use mathematical models with diffusion/reaction equations that capture the known gene regulatory network, to explore the dynamics of these gene expression patterns during early flower development, and to explore whether the final pattern is affected by floral growth or geometry. Unlike in the classical ABC model, our mathematical models suggested that AP1 does not seem to have any major effect on the AG pattern. We then examined published AP1:GFPap1-1 lines and found that in these lines, AP1 expression can be observed in the center of the flower even at later stages. Thus we propose that AG is mainly regulated by AP2 expression.

P-069 From hair to spine: Development of enlarged and asymmetrical awl hair in the spiny mouse (*Acomys dimidiatus*)

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Mammals exhibit highly diversified skin appendages such as the scales of pangolins, the scutes of armadillos and very adapted fur from almost no hair in marine mammals to extremely modified hair follicles becoming spine-producing organs in spiny mice (genus Acomys), hedgehogs and porcupines. New model species are essential to understand the development and evolution of such derived phenotypes. Indeed, these spectacular morphologies have so far not been reproduced in transgenic mice. Here, we undertake a developmental biology and numerical modeling approach to investigate the development of skin appendages in the spiny mouse, *Acomys dimidiatus*. We show that *Acomys* spines are derived awl hairs, one of the four hair types found in the laboratory mouse. *Acomys* spine follicle morphogenesis starts during embryogenesis through interactions between the dermis and the epidermis leading to the formation of an epidermal thickening (placode). Although these initial events are very similar to those observed in the laboratory mouse, *Acomys* placodes are much bigger and the future follicles grow longer and larger. Using tridimensional reconstruction of skin and scanning electron micrographs of spines, we uncover the peculiar morphology of *Acomys* follicles and in particular the highly asymmetrical dermal papilla that sends proliferation signals to the different layers of the forming follicle. Using multiple molecular markers, we demonstrate that the asymmetric shape of the dermal papilla induces two waves of anisotropic growth in the forming follicle that lead to a greatly enlarged matrix at the posterior side of the follicle and a thicker inner root sheath at the anterior side. In combination with keratinization of the cortex and medulla, both anisotropic growths affect the final shape of the spine shaft: the enlarged matrix leads to the formation of a thicker cortex at the posterior side and the inner root sheath compresses the medulla that eventually collapses at the anterior side. Simulations, using linear elasticity theory and the finite-element method, indicate that these processes are sufficient to replicate the time evolution of the *Acomys* spine layers and the final shape of the emerging spine shaft.

P-070 Functional analysis of the FGF ligands FGF8 and Branchless in the Tribolium embryo

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The precise regulation of cell-cell communication by numerous signal-transduction pathways is fundamental for many different processes during embryonic development. One important signalling pathway is the evolutionary conserved fibroblast-growth-factor (FGF)-pathway that controls processes like cell migration, axis specification and mesoderm formation in vertebrate and invertebrate animals. Specifically the ligand/receptor combinations in Drosophila, FGF8(Pyr/Ths)/Heartless(Htl) and Branchless(Bnl)/Breathless(Btl) function separately in regulating the migration of mesodermal cells and in specifying the tracheal network, respectively. In Tribolium only one FGF-receptor (Fgfr) is encoded in the genome. Thus it was hypothesized that both the ligands Fgf8 and Bnl have to signal through the same receptor. We show here that in Tc-fgf8- and in Tc-fgfr-RNAi embryos mesoderm differentiation is impaired. We further show,

that like in Tc-bnl-RNAi embryos the tracheal network has not been differentiated properly in Tc-fgfr-RNAi embryos. By using marker genes for mesodermal and tracheal precursor cells we reveal that the migration of both mesodermal and tracheal precursors require FGFR-dependent signalling. We therefore conclude that in Tribolium and in contrast to Drosophila, a single FGF-receptor integrates the signals of the FGF ligands FGF8 and Bnl during mesoderm- and tracheal development. Our results reveal the plasticity of this important cell-signalling pathway during embryogenesis of animals.

P-071 Functional consequences of lineage-specific duplications: The example of retinoic acid degradation mechanisms in developing Amphioxus

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Retinoic acid (RA) is a small lipophilic molecule that acts as a potent morphogen and has important functions regulating both development and adult homeostasis in vertebrates. Endogenous RA is produced from vitamin A (retinol) in two steps of oxidation and its bioavailability is chiefly regulated by the activity of RA synthesizing and degrading enzymes, respectively called RALDH and CYP26. The signal transduction of RA is mediated by two nuclear receptors, RAR (retinoic acid receptor) and RXR (retinoid X receptor), which act as heterodimers to activate target gene transcription. Although predominantly studied in vertebrates, RA signaling also functions during development of other chordates, including the cephalochordate amphioxus and ascidian tunicates. In contrast, outside the chordate lineage, evidence for RA signaling functions remains scarce. Interestingly, we found evidence that bilaterian genomes typically contain several copies of cyp26 genes, suggesting that different lineages may have independently diversified their repertoire of RA degrading enzymes. The present study specifically aims at revealing the functional consequences of these lineage-specific duplications as well as the evolutionary origin and diversification of RA degradation mechanisms. To this end, we used a combination of *in silico, in situ* and *in vivo* approaches in the cephalochordate amphioxus (*Branchiostoma lanceolatum*) to characterize the expression patterns and functions of the three cyp26 genes encoded in its genome. We found that the amphioxus cyp26 genes are linked in a single genomic cluster and that this organization resulted from tandem duplication events. Quantitative PCR experiments following pharmacological treatments mimicking varying RA levels suggest that the cluster organization is important for mediating a coordinated gene regulatory response to RA signaling. However, the developmental expression of the three amphioxus cyp26 genes indicates that only one of the three paralogs acts as a regulator of RA degradation-dependent patterning during development, while the other two probably function to protect the embryo from RA-dependent teratogenesis. Pharmacology-based experiments on developing amphioxus-using inhibitors of CYP26 activity further support this notion. Altogether this work thus provides a valuable framework for the evolution of novel functions following lineage-specific gene duplication and reveals new insights into the evolutionary history of the RA signaling cascade.

P-072 Gene expression patterns in salamander limb development and their potential role in the evolution of preaxial polarity

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Salamanders display a remarkably different mode of skeletal development in their limbs than other tetrapods. They show preaxial polarity with a sequence of digit developing of II-I-III-IV-(V). All other tetrapods show a postaxial polarity and the sequence of their digit establishment is IV-V-III-II-I. Although the development of the tetrapod limb has been studied for decades, only gene expression and regulatory mechanisms in the limb development of model organism like mice or chicks are well understood today. The differences between limb developmental patterns of amniotes and frogs on the one hand, and salamanders on the other hand are of great interest for understanding the evolution and development of the tetrapod limb. In order to elucidate the developmental basis of the aberrant salamander pattern, we investigate partial and temporal gene expression patterns of genes with well-known roles in tetrapod limb development in the Mexican axolotl (*Ambystoma mexicanum*) and compare these with model organisms like mice or chicks. First results of in situ hybridization analysis of the genes Shh, Bmp-2 and Sox9 will be presented here. Shh is a key regulatory gene of the anterior-posterior axis and preliminary studies focusing primarily on early stages of limb development have shown a much smaller expression area in axolotls than in mice and Xenopus, which could be confirmed in our study. Bmp-2 is considered to be a downstream target of Shh and plays a crucial role in directing digit patterning. While early expression is similar to other tetrapods, divergent expression patterns are detected

in later phases of autopod development. Sox9 is an early marker for chondrogenic condensations stimulated by Bmp-2 and crucial for detecting the early establishment of preaxial skeletal elements. All three genes therefore play an important role in development of the digits and can provide insights into differences of gene expression patterns that may be involved in the coordination of the reversed polarity in the limb development of salamanders.

P-073 Gene trapping in the amphipod crustacean *Parhyale hawaiensis*

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We are carrying out a gene trap screen in the amphipod crustacean *Parhyale hawaiensis* as an unbiased approach to mark diverse cell types and tissues in this organism. These gene traps can be used to study the behaviour of specific cell types during morphogenesis and regeneration, in live animals, and provide a useful platform for functional studies as potential drivers for gene expression. As trapping element we use a transposon vector carrying an exon-trapping construct (a splice acceptor followed by the DsRed coding sequence) and an integrase docking site, allowing for targeted integration of any construct at the trapped locus. Here, we present the outcome of a pilot screen and provide a first characterization of several new traps. We recovered traps marking specific cell types in all three germ layers, which we describe in their distinct expression with respect to their morphology and their temporal expression. We hope that the established transgenic lines will serve to study diverse aspects of morphogenesis and regeneration, and will be of use to a broad research community.

P-074 Genetic and evolutionary basis of sensory diversity

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Evolutionary changes in gene regulatory networks governing development occur either in coding or in *cis*-regulatory sequences. Currently, the "locus of evolution" is under debate. In this project, we study the contribution of variation in transcription factor coding sequence to developmental evolution. To do so, the fruitfly Atonal (Dmato) coding sequence was replaced, in the fly endogenous locus, by seven homologues from distinct animal phyla throughout the metazoan linage (sponges, vertebrates, cephalochordates, annelids, arthropods). Two closely related proteins will serve as controls. The effects of coding sequence substitution on development, morphology and function of the DmAto-dependent sensory organs are under investigation. Eye development is largely unaffected in the different knock in (KI) lines, i.e., in some cases the compound eye is slightly rough but normal-sized. However, in the development of stretch sensitive receptors the different homologs show differences in the number of sensory organ precursor and mature organs. In contrast, once formed, precursors give rise to the proper set of daughter cells, indicating that the sensory lineage itself is not affected. Thus, the homologs differ in their "proneural strength", i.e., in their capacity to specify sensory organ precursors in a tissue specific manner. Currently, we are focused on the molecular basis for the divergence in proneural activity. Experiments to resolve structure/function relationship of the mature protein and to identify protein-protein interaction are initiated. We anticipate that those two approaches will converge. This study provides novel insights into how changes in TF coding sequences affect developmental processes.

P-075 Genetic basis of the evolution of differences in eye size between *D. simulans* and *D. mauritiana*

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In the last decade, the genetic bases for the evolution of particular traits have been identified but, nevertheless, our understanding of the evolution of complex morphological features, and how their underlying genetic changes arose and spread in populations, is still limited. We have found considerable variation in eye size within and among species of the *Drosophila melanogaster* subgroup. In particular, *D. mauritiana* has larger eyes than its sibling species, *D. simulans*, mainly due to differences in ommatidia size. Here, we identify the major loci responsible for the *D. simulans* and *D. mauritiana* eye size differences on the X chromosome by QTL mapping. Using independent introgressions of the QTL region from *D. mauritiana* into the *D. simulans* genome we refined this mapping and restricted it to a region of 1Mb. To further investigate the functional and developmental bases of eye size variation, we performed transcriptome profiling by RNA-Seq of the eye-antenna imaginal discs of the two species at different developmental points. By combining our high-resolution mapping data with our transcriptome datasets, we found differentially expressed genes that lie in the QTL. Finally, we functionally characterized the role of these candidate genes in controlling ommatidia size and number, shedding light into the genetic basis responsible for Drosophila eye evolution.

P-076 Within-species variation in the timing of developmental events: Prevalence, heritability and evolutionary implications

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Natural selection acts on all stages of development, yet our understanding of the extent of variation present during early development and the evolutionary implications of such variation is limited. Using embryos of the pond snail *Radix balthica* we have investigated variation in the timing of a suite of both morphological (e.g., shell formation, eye formation) and physiological (e.g., crawling, first heart function, hatching) developmental events that occur at particular times during the embryonic period. This variation is substantial and greatest at the inter-individual rather than inter-clutch or inter-population level, even when comparing populations from the extremes of this species' range. Within a single population inter-individual, genetic differentiation is a good predictor of the degree of dissimilarity in the timing of a number of developmental events, suggesting an underlying genetic basis for differences in developmental event timings. Furthermore, a comparison of the timing of developmental events in the embryonic stage in parents and their offspring demonstrated that the timings of foot attachment and first crawling were heritable. We suggest that inter-individual variation in developmental event timing may well be the raw material from which some inter-specific heterochronies originate, via natural selection.

P-077 Genomic analysis of life cycle evolution in Culex mosquitos

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Developmental pathways are highly conservative in most animal phyla, but may be involved in local adaptations. Common house mosquito *Culex pipiens* is a widespread disease vector forming many local populations in various habitats. Urban populations (biotype molestos) have very specific life traits: they can lay eggs without blood sucking, mate in small chambers and do not diapause. To find the possible genetic basis of life cycle differences between urban and natural populations of *C. pipiens* we resequenced pooled samples of six populations from urban and natural habitats and two populations of the outgroup *C. torrentium*. Principle component analysis of Tajima's D and selective sweep scores revealed that although the majority of genes experienced concordant selective pressure across Culex species and populations, „differential selection" caused the samples to cluster according to localities not biotypes or habitats. Multiple adaptive events, involving genes implicated with regulation of autogeny, diapause, insecticide resistance and heat shock response were limited to specific populations. We can hypothesize that both the autogeny and lack of diapause can be induced by disrupted Notch-pathway, and we found it under positive selection in urban populations. About 5-20% of the genes and almost half of the annotated pathways were under positive selection in each population. Genes, involved in many pathways, demonstrated positive selection in various populations. Introducing the new quantitative definition of pleiotropy as the number of biological pathways in which a gene in involved (based on KEGG annotations) we showed than having several functions did not limit the gene potential for adaptive evolution. The adaptive importance of regulatory changes in gene expression is supported by high occurrence of sweeps in nongenic regions and in chromatin-remodeling genes. Histone H1genes experienced parallel directional selection driven partly by recent proline mutations.

P-078 Geometric morphometrical analysis of the evolutionary development of carnassial teeth in extant canids

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In this study we analyzed the form of the lower carnassial tooth of 33 extant species of the subfamily Caninane (Canidae, Carnivora). We related the carnassial form to different dietary habits and taxonomic genera. We analyzed the complete carnassial outline, and the talonid and trigonid regions separately. We used Elliptic Fourier Analysis (EFA) to describe the profile of different parts of the carnassial tooth. To be able to observe how the data groups together we did principal component analysis (PCA), we also used Canonical Discriminant Analysis (CDA) to analyze the distribution in relation to the first two principal components of the different groupings. The explained variance of the first two principal components was always above 80%. A high percentage of individuals was classified correctly for the guilds as well as the genera in all analysis. We conclude that alimentary habits had strong correlation with the form of the carnassial tooth. in conformational space, the conformation associated with carnivory is located in a middle position between omnivory and hypercarnivory. We could also observe a high proximity in shape space among South american genera. At the same time we could observe that insectivory in canines is represented by different carnassial shapes that could have evolved independently.

P-079 Germline stem cells and cluster formation in the polytrophic meroistic ovary of *Nasonia vitripennis* (Hymenoptera)

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Many animal species owe their often life-long reproductive capacity to the presence of germline stem cells (GSCs) in their gonads. So far, research on GSCs has been focused on a few model organisms, among them Drosophila. In the meantime, many different aspects of GSC biology are reasonably well understood in the fly and especially the ovarian GSCs of Drosophila still serve as an important model for the regulation of adult stem cell behaviour *in vivo*. In the polytrophic meroistic fly ovariole 2-3 solitary GSCs adhere to a somatic niche that governs GSC maintenance, function and differentiation mainly via paracrine signaling. GSCs self-renew by asymmetric mitotic divisions that give rise to one daughter cell that maintains its GSC character and another daughter cell that leaves the niche and differentiates into a so-called cystoblast. Each cystoblast undergoes four rounds of synchronous mitoses followed by incomplete cytokineses thereby producing a cluster of 16 interconnected cystocytes, one future oocyte and 15 sibling nurse cells. It is widely accepted that Drosophila represents an insect group with highly derived evolutionary traits. Thus, it is questionable to what extent the fly ovary is applicable as a model for the polytrophic meroistic insect ovary in general. The polytrophic meroistic ovary type, however, is believed to represent the ovary of the last common ancestor of all holomentabolous insects. Among these the hymenopterans are classified as the basal most taxon. Considering these facts, we chose the hymenopteran *Nasonia vitripennis* to start a functional study of the early processes of polytrophic oogenesis in a basal holometabolous insect. Like in Drosophila, maturing *Nasonia* germ cell clusters consist of one oocyte and 15 nurse cells. *Nasonia* females exhibit a lifelong capacity to produce eggs, which we could show is fueled by germline proliferation during the whole lifespan of adult *Nasonia* females. Our immunocytochemical and ultrastructural analyses revealed a GSC-like population of mitotically cycling, small germ cell clusters in the anterior portion of the *Nasonia* germarium and — as far as possible — we could rule out the existence of solitary GSCs. RNAi mediated knockdown of the intrinsic maintenance factor nanos in Nasonia led to a loss of this anterior most, mitotically active population of germ cell clusters by differentiation. This result strongly indicates a conserved function of Nv-nanos for the maintenance of the undifferentiated state of these clusters and reflects their stem cell-like character and function. Knockdown of Nv-pumilio, an interaction partner of Nanos in Drosophila GSCs, on the other hand led to largely non-overlapping defects in this region, suggesting a Nv-pumilio independent function of nanos for GSC maintenance in Nasonia. Summing up, we propose a model for GSC function in Nasonia ovarioles in which an anterior population of small, undifferentiated germ cell clusters retains self-renewing potential that depends on Nv-nanos function.

P-080 Gonad regeneration in medusae of the hydrozoan *Clytia hemisphaerica*

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The phylum Cnidaria displays a great diversity of life cycles and modes of reproduction. During the life cycle of the hydrozoan experimental model *Clytia hemisphaerica*, a colonial polyp asexually generates short-lived medusae, which develop gonadal structures, containing male or female gametes (Houliston et al. 2010). This mode of reproduction, deeply different from the ones of most studied bilaterians, poses interesting questions related to the localization and the regulation of the stem cells employed in the production of new individuals. Stem cells are also implicated in regeneration of lost parts of the colony, and of the medusa. The remarkable regenerative capacities of cnidarian polyps have been studied in species such as *Hydra* and *Nematostella*, however, only few pioneering studies have focused the medusa stage (Schmid & Tardent 1971). *Clytia* medusae normally develop four gonads, positioned on the four radial canals running between the manubrium (mouth) and one of the tentacle bulb located on the bell margin. We are investigating the mechanisms that control the precise positioning and differentiation of the gonads during ontogeny and regeneration, from an initial aggregate of undifferentiated cells to the final structure composed of germ cells and somatic cells (from ectodermal and endodermal epithelia). We show that *Clytia* medusae possess extensive regenerative capacities, and are able to reform very rapidly both germinal and somatic parts of the gonad. We have found that if the radial canals are completely excised with the gonad, they regrow both from the manubrium and from the tentacle bulb, fuse and a new gonads appear at the place of the excised one. This process likely involves both differentiation of stem cells and de-differentiation of umbrella cells, as well as signaling between the two growing canal tips. We are currently performing transcriptome and functional analyses to compare ontogenetic and regenerative processes of gonad formation. This will allow us to investigate the regulation of stem cells and germ stem cells in a simple system, which still employs the

transcription factors and signaling pathways described in Bilateria, but which displays an exceptional capacity for reprogramming in the context of regeneration.

P-081 Growth vigour of genotypes with impaired and enhanced S-nitrosothiol signaling indiates nitric oxide feedback regulates assimilation in Arabidopsis

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Plants must acquire nutrients from soil in order to complete their life cycle. Nitrogen assimilation constitutes a particularly limiting factor in plant development and crop yield. The primary source of nitrogen to many plants is inorganic nitrate. Once taken up by roots nitrate is reduced to nitrite by the activity of NAD(P)H-dependent cytosolic Nitrate Reductases (NR). Nitrite, in turn, is promptly removed from cells or transported to chloroplasts where it is reduced to ammonium for further assimilation into organic compounds. In addition, nitrite can be reduced to nitric oxide (NO) via non-enzymatic as well as various enzymatic pathways. NO is a free radical that acts as a redox signal in eukaryotes. The addition of NO to cysteine residues in proteins results in the formation of S-nitrosothiols (SNO), which have been shown to alter the activity, localization, and conformation of target proteins. NO may also react with glutathione to form S-nitrosoglutathione (GSNO), a major cellular reservoir of bioactive NO. Cellular GSNO levels are controlled by the evolutionary conserved, cytosolic enzyme GSNO Reductase 1 (GSNOR1). Here, we demonstrate genetic and biochemical evidence for intimate interplay between nitrate assimilation and NO signalingin the model plant *Arabidopsis thaliana*. To assess the physiological impact of (S)NO on nitrate assimilation, we analysed the vigour of (S)NO signalling mutants by measuring growth and biomass accumulation parameters. As expected, inability of NR knockout mutant plants to reduce nitrate led to a decrease in leaf area and dry shoot weight compared to WT. Like NR mutants, nitric oxide overproducer1 (nox1) and gsnor1 deficient mutant plants also displayed strongly decreased growth vigour. On the other hand, leaf area and biomass accumulation tended to increase, in GSNOR1 overexpressing 35S::FLAG-GSNOR1 plants. Importantly, irrigation of nox1 and gsnor1 mutants in the presence of glutamine, the main end product of nitrate assimilation, recovered growth vigour of gsnor1, but not that of nox1, to levels comparable to those of WT and 35S::FLAG-GSNOR1 plants. Accordingly, measurement of enzymatic activity revealed that NR is negatively impacted in gsnor1 mutants, but not in nox1, compared to WT plants. Conversely, GSNOR1 overexpressing plants displayed the highest NR activity among the genotypes, suggesting that GSNO and free NO differentially affect NR activity. Taken together, our data illustrate that (S)NO are important regulators of nitrate assimilation and, consequently, control plant growth and development.

P-082 Halisarcidae (Demospongiae) ectosome regeneration: Mesenchymal morphogenesis and epimorphosis

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The data on mechanisms of morphogenesis must be accumulated and compared between sponge lineages and eumetazoans in order to understand the early evolution of animal pattering in relation to genome evolution. Sponges are key group to provide significant answers to these fundamental evolutionary questions. For this purpose, we have combined several techniques to study mechanisms of regeneration of the ectosome in non-skeletal sponge *Halisarca dujardini* (Demospongiae). There are three main steps of regeneration: (1) formation of „the regeneration plug" (6 - 12h), (2) wound surface epithelization (24 - 36h), (3) the end of regeneration - the regenerate presents a functional exopinacoderm and aquiferous system canals (48 - 72h). At 12h after injury, the wound surface begins cover by extracellular matrix. At 24h, the wound is covered by a dense collagen layer. Cells migrate individually to the regenerate surface. Separated exopinacocytes begins to differentiate individually without formation the epithelium. There is no any movement of intact exopinacoderm around the wound. Cells of the mesohyl are filled with the phagosomes. By 48h the wound is covered by exopinacoderm, choanosome begins reconstruct. In *H. dujardini* there are three main sources of exopinacoderm: archaeocytes, choanocytes and, rarely, endopinacocytes. For DNA synthesis labeling with 5-Ethynyl-2'-deoxyuridine (EdU) was used in final concentration 800 uM. Cells (mainly choanocytes) in intact sponge actively inco0rporate EdU. During regeneration the most actively DNA synthesize cells are choanocytes that migrate at the wound surface and stops or slows down their cell cycle, whereas the choanocytes in underlay tissues continue replication. No any local proliferation at wound surface or in deeper layers of mesohyl was detected. We sequenced transcriptome and genome in *Halisarca* and looked for Wnt genes. Ten Wnt genes were identified, and at least four of them involved in apical-basal polarity in *Halisarca*. One of Wnts (HdWntK) expresses in narrow pattern at the edge of wound between 6 and 24h of regeneration. The regeneration in *H. dujardini* is an epimorphosis that requires active cellular proliferation and dedifferentiation prior to the replacement of the lost body part. This regeneration combines two mechanisms

to form the regenerated structure: cells dedifferentiation followed by redifferentiation and the intervention of stem cells. Mesenchymal morphogenesis by mesenchymal-epithelial transformations is the main mechanism during *H. dujardini* regeneration.

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P-083 Heads and tails: To be or not to be?

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Polychaetes vary greatly in their regeneration abilities. Even among representatives of one family there are species that can regenerate both anterior and posterior parts of the body, those that restore only posterior part or don't regenerate at all. This makes the polychaetes a perfect model for studing the mechanisms of regeneration that are still poorly understood. In our previous work we described the activation of Hox gene system during the regeneration of *Alitta virens*. Although this polychaete can successfully restore the posterior part of the body it lacks the ability to regenerate the "head". One of the earliest events during *A. virens* regeneration is the activation or repression of the number of Hox genes near the amputation site (Novikova et al. 2013). We believe that this expression marks the new posterior end of the worm's body and plays an important role in the regeneration process. Using WMISH we investigated the behavior of some Hox genes in the posterior parts of the worms' bodies after dissecting the worm into two halves. Surprisingly, we observed the activation of some "posterior" Hox genes near the amputation site of the "tail" part of the body from where the head is supposed to grow out. By marking this territory as the posterior one Hox genes may prohibit the head regeneration of *A. virens*. This mechanism may partially explain the absence of head restoration in some polychaete species.

P-084 Heterochronies in teleost caudal fin evolution: Experimental evidence

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The caudal skeleton of fish is one of morphological systems with great phylogenetic, ontogenetic, and functional significance. The caudal system shows a great morphological disparity, resulting from ontogenetic and phylogenetic losses and fusions of various skeletal elements (hypurals, epurals, vertebral centra) during the evolution of teleosts. One of the proposed mechanisms for evolutionary changes in fish skeletal system is heterochrony, changes in the developmental rate and timing, leading to changes in adult morphology. To verify role of heterochronies in the evolution of fish caudal complex, we artificially modified the rate and timing of the ontogeny events of several fish species, and assessed the consequences of these changes for the morphology of caudal system. Temporal changes in ontogeny were caused by alterations in the level of thyroid hormones, the main regulators of fish development. The results of the experiments show that artificially induced heterocronies resulted in changes of the number and size of caudal system elements: vertebral centra, hypurals, epurals, fin rays. This finding indicates participation of heterochronies in the caudal complex evolution.

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P-085 How to build an ectodermal organ? Inferring the minimal gene networks able to generate different types of ectodermal buds

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The morphogenesis of animal organs and structures is driven by the coordinated growth and movement of cells and tissues. The gene networks controlling these processes usually have a more complex topology compared to the theoretically simplest network capable of doing it. This makes the integrative understanding of those networks and the comparison between different systems more difficult. Thus, inferring the minimal gene network able to direct the development of an organ can be a powerful tool to understand the overall logic of the process and will allow to compare shared aspects and differences with other systems. Ectodermal organs, despite showing a wide range of different forms and structures, originate from the same type of structure, the ectodermal placodes, and their development is regulated by the same set of signal transduction pathways. In our study, we calculate the minimal gene network capable of directing the development of different types of ectodermal buds using a 3D computational model that simulates the developmental dynamics of epithelial-mesenchymal tissues, gene network dynamics and extracellular signal diffusion. We use the developmental model to test which gene networks are capable of forming ectodermal (evaginating or invaginating) buds by allowing certain genes to affect different cell behaviors like cell division, cell migration and cell adhesion among others. We use first an evolutionary algorithm to search for networks capable of generating buds by selecting for a certain bud morphology. Then we use a second reductive algorithm on those networks in order to simplify them. We delete one connection from the network and see if it still generates a bud. If this is not the case, the connection is

restored and another one is selected for removal. Our results may shed light into the overall logic of the gene networks directing ectodermal organ morphogenesis and allow to identify shared features between different types of ectodermal organs that may account for conserved developmental modules.

P-086 Hyoid first: Heterochronic development of bichir hyoid metamere

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Bichirs (Polypterus) include the basalmost representatives of extant actinopterygian fishes and therefore are uniquely well suited for assessing the ancestral character states of Osteichthyes and Actinopterygii, as well as the divergence of developmental patterns between, e.g. fishes and tetrapods. Bichirs posses a unique set of features among which external gills are of special interest, since they constitute the first organ visible on their head. Precocious formation of this key larval adaptation — which in bichir is situated only on the hyoid arch — is crucial for their survival in less oxygenated water. Our analysis of bichir development revealed a striking heterochrony shift in formation of the entire hyoid region or metamere. This primarily includes accelerated migration of hyoid neural crest cells: this stream is more abundant and migrates much earlier when compared to other neural crest streams, unlike in other vertebrates. Mesenchyme from the hyoid neural crest stream contributes into voluminous supporting tissues of bichir external gills. Much sooner migration of neural crest cells is developmentally allied with a very early lateral expansion of the second pharyngeal pouch endoderm that is responsible for morphogenetic formation of bichir external gill anlage. Formation of the hyoid aortic arch, supplying the oxygen for external gills, is highly accelerated too. Our data reveal that very early formation of bichir external gills is developmentally driven by heterochronic acceleration of multiple germ layers of bichir hyoid metamere.

P-087 Identification of a preformed germ plasm in the sexual oviparous pea aphid: A non-canonical case in the Hemimetabola

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Germline specification in some animals is driven by the maternally inherited germ plasm during early embryogenesis (inheritance mode), whereas in others it is induced by signals from neighboring cells in later embryogenesis (induction mode). In the Metazoa, it has been clear that the induction mode is the more prevalent and ancestral condition; the inheritance mode is therefore derived. For example, in Drosophila and several other holometabolous insects such as mosquitos and wasps, germline specification follows the inheritance mode. By contrast, in most hemimetabolous insects like crickets and grasshoppers, germ cells are segregated after blastoderm formation via signal induction. Although the inheritance mode is not common in the Hemimetabola, a putative germ plasm has been identified in the mullein thrips *Haplothrips verbasci* — a member of the order Thysanoptera — using traditional microscopic approaches. This implies that some insects belonging to the superorder Paraneoptera — a sister group to the Holometabola that includes orders Psocoptera, Phthiraptera, Thysanoptera, and Hemiptera — adopt the inheritance mode to specify germ cells. However, no molecular evidence could support such hypothesis before we studied germline specification in the sexual oviparous pea aphid *Acyrthosiphon pisum*, a hemimetabolous insect belonging to the Hemiptera. We employed Apvas1, a *Drosophila vasa* ortholog in the pea aphid, as a germline marker to examine whether a preformed germ plasm is assembled in the sexual pea aphid. Using an antibody against ApVas1, we identified a preformed germ plasm in the syncytial blastoderm and later we could trace its specific expression in the primordial germ cells (PGCs). Expression of Apvas1 mRNA was not restricted to the presumptive germ plasm in uncellularized embryos whilst co-localization of Apvas1 mRNA and ApVas1 protein could be detected in PGCs later throughout embryogenesis. To our knowledge, this is the first case showing the inheritance mode being adopted by the paraneopteran insects at molecular level. Compared with the induction mode identified in other hemipteran species such as the milkweed bug *Oncopeltus fasciatus*, our study suggests that formation of the preformed germ plasm in the pea aphid is achieved via independent evolution in the Hemiptera.

P-088 Identification of MADS-box genes of the AP1/FUL clade in *Passiflora edulis* (Passifloraceae)

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The flowers of the genus Passiflora are noteworthy by their extraordinary diversity in colors and shapes. Some floral features, such as the corona filaments and the androgynophore are family-specific. Additionally, most of the Passiflora are climbing vines in which tendrils are thought to be a modified flower. The evolutionary basis that gives rise to such floral structures is not clear yet. The MADS-box transcription factors from the AP1/FUL lineage have been shown

to be key regulators of the floral meristem and perianth identities and therefore are potential candidates to play roles in Passiflora floral development. In this work we identified potential orthologs of AP1 and FUL in *Passiflora edulis* using EST and RNAseq databases, and named them PeAP1 and PeFUL respectively. PeAP1 encodes a predicted protein of 243 aminoacids while the putative PeFUL has 241 aminoacids. A phylogram using PeAP1, PeFUL and other representative protein sequences of AP1/FUL clade suggested that PeAP1 belongs to the known euAP1 clade and PeFUL to the euFUL clade. Furthermore, a comparative analysis of the protein sequences showed that PeAP1 shares the two euAP1 conserved terminal motifs, the farnesylation signal and the transcription activation domain. On the other hand, PeFUL shares the euFUL and FUL-like motif that comprises six hydrophobic aminoacids. The results of expression analysis of these genes in different tissues pointed to their possible roles in contributing to the morphological diversity in the genus.

P-089 Identification of new dorsoventral patterning genes by differential expression analyses in *Tribolium castaneum*

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The BMP pathway plays a conserved role during the evolution of almost all bilateria. It is a major factor in dorsoventral (DV) patterning of vertebrates, but also in the arthropode lineage. However, during insect evolution Toll-signaling took over functions from BMP-signaling pathway in DV patterning. In the highly derived group of Drosophilids, dorsoventral patterning is dominated by Toll-signaling, whereas BMP-signaling has a limited function downstream of Toll. Extensive studies have led to a thorough understanding of the mechanisms that act during dorsoventral patterning in the fruit fly *Drosophila melanogaster*. However, little is known about the gene regulatory network (GRN) that acts during establishment of the dorsoventral axis of more basally branching insects like the red flour beetle Tribolium. Although they interact differently, the transcription factors Dorsal and Twist initiate DV patterning in Tribolium and Drosophila and likely control major parts of the DV GRN in both organisms. We identified potential target genes for Tc-Dorsal and Tc-Twist, but also for the more dorsally acting DV components Tc-Sog and Tc-Dpp in a global and unbiased manner. To achieve this, a transcriptome analysis via RNA-sequencing was performed. To identify differentially expressed genes, the transcriptomic data of knockdown embryos were compared to control samples. Furthermore, we analyzed the overlapping up-and/or down-regulated genes of Toll-RNAi vs. twist-RNAi and sog-RNAi vs. dpp-RNAi. In addition, putative Dorsal binding sites were identified by ChIP-sequencing. The combination of both analyses resulted in a list of genes either regulated by Dorsal, by twist or by both. An in-situ hybridization screen was performed to reveal mRNA expression patterns of these bona fide DV patterning genes during early Tribolium embryogenesis. Furthermore, genes with interesting expression patterns in blastoderm or germband stages were chosen for further functional studies by means of pRNAi. By this approach we were able to identify further genes involved in the DV-GRN of *T. castaneum*. Functional studies of the investigated candidate genes will lead to a better understanding of the evolution of the DV-GRN in insects.

P-090 Identification of target genes of the terminal system in *Tribolium castaneum* by next-generation-sequencing

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In the long-germ insect Drosophila, the so-called terminal-system operates during early stages of embryogenesis to specify terminal cell fates. Failure to activate Torso-signaling results in defects in head formation and loss of posterior segments posterior abdominal segments. Previously, we showed that in the short-germ beetle Tribolium Torso-signaling is required for setting up or maintaining a functional growth zone and, at the anterior, for the extraembryonic serosa. However, while Torso-signaling in Tribolium provides anterior and posterior polarity cues, the target genes of the terminal system are largely unknown. In order to uncover additional target genes of the terminal system in Tribolium, we performed a differential gene-expression-screen, using SOLiD Next Generation sequencing. We sequenced transcriptomes of 7-12h wildtype, torso-like (tsl) RNAi, and capicua (cic) RNAi embryos. The HMG-box protein Capicua acts as an antagonist of the terminal system and thus, the depletion of cic results in the de-repression of terminal target genes. By comparing loss-of-function (i.e. tsl RNAi) with gain-of-function (i.e. cic RNAi) transcriptomes, we identified more than 500 differentially expressed genes. To elucidate whether these genes are indeed (directly or indirectly) associated with terminal patterning, we conducted a mini-screen for 50 of the most interesting candidates by parental RNAi. In addition, we monitored expression of candidates and tested for marker gene expression. Strikingly, 80% of these candidate genes are involved in early anterior and/or posterior pattern formation, indicating that en mass sequencing indeed can identify novel target genes of the terminal system in Tribolium.

P-091 Identifying the developmental genetic basis of craniofacial evolution using Threespine Stickleback

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The developmental genetic basis of adaptation to novel environments is still poorly defined. We are studying threespine stickleback fish as a means to better understand how rapid adaptation at the genomic level can translate into phenotypic differences through modifications of development. Tremendous change in head shape morphology has accompanied the repeated and independent invasion of oceanic threespine stickleback into freshwater habitats. A functionally important bone in the head of fish is the opercle, which is a key component of the structure that covers the gills and is essential for feeding and respiration. Evolution of the opercle bone shape is particularly important for adaptation to novel environments because of the profound effects on feeding mechanics. Much has been learned about the development of the opercle in zebrafish, but little is known about what genes underlie opercle shape change in natural populations, or how variation in those genes alter developmental processes to give rise to opercle shape changes in stickleback evolution. We are using 50 year-old populations of Alaskan stickleback to identify genes associated with OP shape change through genome wide association. Furthermore, we use lab populations of Alaskan oceanic and freshwater stickleback with differentiated opercle shapes to identify differences in bone outgrowth, cellular behavior, and gene expression through development that may contribute to opercle shape change.

P-092 Independent evolution of snakelike morphologies in Squamata: Do lineages with equivalent morphologies share molecular signatures in developmental genes?

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Snakelike morphologies consist of elongated trunks and reduced and/or absent limbs. This type of morphology evolved multiple times within Squamata (Tetrapoda), being two classical examples of snakelike animals: the amphisbaenians and the snakes. The axial morphology of these two lineages also has a peculiar pattern: all trunk vertebrae are associated to ribs. Given that the snakelike morphology evolved independently in the Serpentes and the Amphisbaenian lineages, these evolutionary transitions potentially involved changes in developmental pathways that are exclusive to each clade. However, developmental programs have been described as being conserved among vertebrates, and perturbations in development often trigger pleiotropic effects. It is possible, then, that the independent evolution of snakelike morphologies in Serpentes and Amphisbaenians also imprinted similar molecular signatures in these lineages. We sequenced fragments of coding (Hoxa13) and non-coding (CsB and H1-Myf5) regions of genes expressed during embryo development, and compared their molecular signatures between Serpentes and Amphisbaenians. Analyses of coding regions compared models of molecular evolution between lineages using PAML, while non-coding regions were compared between clades based on predicted transcription factor binding sites (TFBS). Our analyses identified in the three fragments of snakes some molecular signatures that are absent in amphisbaenians. The most striking one is a polymorphism in H1-Myf5 described in the literature as being responsible for lack of identity in trunk vertebrae of snakes that is absent in amphisbaenians. The analyses based on prediction of TFBSs not only reveal regulatory haplotypes in CsB and H1-Myf5 that are exclusive to Serpentes or to Amphisbaenians, but also show one substitution (replacement of a binding site for HIC1 by one for PBX1 in the studied CsB fragment) that is observed in both lineages. Our results support that independent evolution of snakelike morphologies in Squamata imprinted molecular signatures that are exclusive of each lineage, but we also found evidence for possible regulatory convergence in specific regions, which may indicate some degree of conservation in the way developmental pathways can be modulated during these evolutionary transitions.

P-093 Inferring chewing motion and development from adult dental morphology

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The evolution of mammalian dentition is constrained by functional necessity and by the non-independence of morphological structures. Efficient chewing implies coherent tooth coordination from development to motion involving covariation patterns within dental parts. These covariation patterns can be detected using geometric morphometrics. Morphological traits sharing strong covariation (integration) can thus be defined as modules. Integration patterns between and within the upper and lower molar rows are

analysed to identify potential modules and their origins (functional and developmental). To explore patterns and processes at both developmental and functional levels, dental innovation in arvicoline rodents is a case of interest. They exhibit a highly derived molar phenotype with a complex and flat occlusal surface composed of alternate cusps. The number of cusps varies both for the teeth on a row and between the upper and lower molar rows producing a particular form of occlusion between opposing molars. Results support an integrated adult dentition pattern for both developmental and functional aspects. The integration pattern between opposing molar pairs suggest a transient role for the second upper and lower molars during the chewing motion. One of the main conclusions is that the relative integration of molar pairs observed in adults is in contradiction with existing developmental models. Integration produced by late developmental mechanisms may hide the early integration. Emphasis only on the first three cusps to grow leads to a covariation pattern congruent with developmental models. Careful attention to interesting features brought to light by developmental studies makes adult morphology relevant for the interpretation of modularity and integration patterns at both functional and developmental levels.

P-094 Integrating phylogenetics, ecology and evo-devo to understand the origin of plant species: The role of spur length evolution in speciation of the genus Linaria

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Understanding the origin of species is one of the major challenges of modern biology. It requires the integration of ecological, evolutionary and developmental approaches. In this project, we aim to understand speciation processes in plants using a clade of eight Iberian bifid toadflaxes (Linaria sect. Versicolores) as a model system, and applying a multidisciplinary approach. Particular attention is paid to investigating the role of evolutionary variation in the length of nectar spurs in mediating speciation, as nectar spurs have been previously hypothesized as a key innovation promoting speciation in angiosperms. Our objectives are: (1) To reconstruct phylogenetic relationships within the study clade using multiple unlinked DNA regions and coalescent-based methods. Preliminary results indicate a close relationship and recent divergence (<0.5 Mya) between species with highly divergent spur lengths *(L. clementei*, c. 3 mm; *L. salzmannii*, c. 13 mm), suggesting a role of spur length variation in speciation. (2) To investigate the components of reproductive isolation among species, including prezygotic (pollinators, environmental niche) and postzygotic breeding barriers. The species we study are pollinated by insects (mainly bees) with varying proboscis lengths that are potentially involved in prezygotic isolation. Postzygotic barriers between closely related species are considered weak in *Linaria*. (3) To understand the genetic and developmental basis of spur length variation. To this end, we will build upon previous work suggesting a role of KNOX genes in spur development in *Linaria*. We will analyze the transcriptional basis of spur development in *Linaria* using next-generation sequencing technology. Then, inter-specific differences in expression patterns of genes potentially involved in regulating spur length variation will be examined. In the end, information on phylogenetic relationships, reproductive barriers, evolution and development of key traits will be integrated in order to understand mechanisms of speciation in Linaria sect. Versicolores.

P-095 Investigating complex leaf development with the Bladderwort *Utricularia gibba*

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Previously, we published a model that describes the development of the Arabidopsis leaf (Kuchen et al. 2012). This model demonstrates how polarity across the leaf tissue can provide axial information, enabling local specification of growth rates parallel and perpendicular to this axis. A key question is whether the principles employed by this model can be used to explain the generation of diverse forms or whether more complicated mechanisms may be at play. Epiascidiate (cup-shaped) leaves of pitcher plants are one of the most complex leaf shapes in nature. Interestingly, this shape has evolved four times independently in the families Nepenthacea, Sarraceniaceae, Cephalotacea (pitcher plants) and Lentibulariaceae (bladderworts). This may indicate that relatively simple changes in an underlying developmental system may have taken place to give rise to this repeated evolution of form. To determine whether this may be the case, we are exploring how epiascidiate leaves develop in the bladderwort *Utricularia gibba*. We are working to establish *U. gibba* as a model plant and we are investigating the development of its epiascidiate bladders using a combination of imaging, molecular-genetic studies and computational modelling. This should reveal whether epiascidiate leaf morphogenesis may be accounted for by similar principles to those described for the Arabidopsis model.

P-096 Is the successional dental lamina initiated in species with one generation of teeth?

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Successional lamina plays an essential role in the development of replacement teeth in diphyodont and polyphyodont animals. However, it can be also found in monophyodont species where only one generation of teeth is initiated. Here, we aim to analyze morphological changes of the successional lamina development in mouse during embryonic and postnatal stages. Successional lamina of the mouse was morphologically recognizable in the area of the first molar at the bell stage (E14). The lamina was very short finger-like epithelial protrusion, which was formed on the lingual side in the close proximity to the outer enamel epithelium. At E18, folds of the basal membrane were noticeable at the tip of the lamina. Cells attaching to the basal membrane had a cylindrical shape with oval nucleus and smooth nuclear membrane. Cells were in a close contact to each other. Mesenchymal cells surrounding the successional lamina were flattened and concentrically arranged around the finger-like protrusion. During the postnatal stages, the successional lamina became thinner and shorter. Furthermore, it turned to be more tightly connected to the tooth and hardly observable from the outer enamel epithelium. We observed successional lamina until the stage P10 where only a small projection was seen at the base of the dental stalk. Only a few TUNEL-positive cells were found in the successional dental lamina through its regression. Successional lamina was PCNA-negative at postnatal stages in agreement to its decreasing size during these stages. Therefore, the apoptosis does not seem to be a main process of successional lamina regression in the monophyodont mouse but the reduction of successional dental lamina size is probably caused by the inhibition of proliferation, which led to a decrease of cell population during postnatal stages and final failure of replacement tooth formation.

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P-097 Left-right asymmetric control in hemichordate acorn worm embryos

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Consistent left-right (LR) asymmetric organogenesis is a common feature in bilaterians. In all the chordate species studied, nodal, which encodes a transforming growth factor β ligand, is consistently expressed on the left side. Nodal signal then triggers a left-sided transcriptional cascade and activates expression of two downstream genes, lefty and pitx2. The Nodal-induced transcriptional cascade is also conserved in several nonchordate animals. However, in sea urchins, Nodal signaling is activated on the right side to prevent formation of the adult rudiment. It is unclear whether the right-sided nodal expression is an echinoderm innovation or a conserved nonchordate deuterostome character. To address this question we study the LR asymmetric control in hemichordates, the sister group of echinoderms; together they are referred to as the Ambulacraria, which are closely related to chordates. In this study we show that in the hemichordate acorn worm embryo, nodal, lefty, and pitx2 are all expressed on the right side. The right-sided gene expression is perturbed when Nodal signaling is inhibited. Moreover, similar to sea urchins and several chordate species, the LR asymmetric gene expression is randomized in H+/K+-ATPase inhibitor-treated acorn worm embryos. Our findings thus suggest that the right-sided Nodal signaling cascade has been utilized in the Ambulacraria ancestor and that H+/K+-ATPase is involved in LR asymmetric controls in all three major groups of deuterostomes.

P-098 Localization of beta1 integrin and fibronectin during mussel development

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The β1 integrin subunit is considered to be the most conservative among all known β integrins and suggested to be their common ancestor with an important role in embryogenesis and tissue morphogenesis in vertebrates and invertebrates. However, there are no clear data on the localization of β1 integrins in the development of Mollusca, one of the most successful metazoan phyla. We investigated the contribution of β1 integrin to mussel larval morphogenesis, including the development of the digestive system and muscle and neuronal differentiation. We found that the first β1-like integrin positive signal appeared in the forming stomach area at the early trochophore stage. Later, β1-like integrin was localized in cell membranes of the larval stomach. We used a double immunostaining followed by confocal microscopy for simultaneous detection of β1-like integrin and myosin (marker of muscle cells) or serotonin (neuronal marker). Neither muscle cells nor neuronal cells expressed β1-like integrin. In addition, we monitored the expression pattern of fibronectin (FN), one of the major molecules of vertebrate matrices and an ECM-ligand of integrin receptors containing β1-subunit, from

fertilized eggs until the veliger stage. First FN-positive cells were detected at the blastula stage without any β1-like integrin-positive cells present. Later, FN-positive cells were located primarily in the velum of veligers and in the cells surrounding the stomach. However, no co-localization of FN with β1-like integrin was observed throughout early mussel development, indicating a lack of interaction between these proteins. Thus, our results show that β1 integrin can be proposed for use as a specific marker of digestive system in Mytilus larvae.

P-099 Mapping ephippia color variation in *Daphnia magna*

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Colour variation is commonly present in nature. It is usually a complex trait, both in the factors driving it and in the genetic architecture underlying it. In Daphnia, previous studies have shown that body colour varies due to the plastic response of a single gene to the environment. In this project, we study for the first time the colour variation in the ephippial eggs of the freshwater crustacean *Daphnia magna*. After following several clones form different populations in the laboratory we observed that each clone produced consistently ephippia with the same colour. Specifically, we observed that some clones produced black ephippia while others produced brownish ephippia. By using a QTL panel with parents that produce different ephippia colours we were able to map this trait. The analysis of this QTL shows that ephippia variation colour is likely determined by two main loci and the genetic component responsible for the black phenotype is recessive. Furthermore, our QTL analysis also identified a third locus that despite the genotypes at the first two loci is able to switch the final phenotype of the ephippia. We speculate that this third locus acts as a developmental switch for color variation in response to environment.

P-100 Mapping the corn snake pigmentation and colour pattern mutations using NGS technology

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The corn snake (*Pantherophis guttatus*) is an emerging model for studies of the genetic basis of pigmentation and colour pattern formation in reptiles. Indeed, selective breeding and spontaneous mutations in that species have generated a variety of colour and pattern phenotypes. Here, we used NGS sequencing together with a linkage mapping approach to unravel the genetic basis of several alleles that affect either melanin-based pigmentation or colour pattern formation. We generated mapping panels of nearly 200 individuals segregating for the target alleles. We used subsets of these individuals (i.e., pools of 20 mutant and 20 wild-type offspring) to genotype single-nucleotide polymorphisms (SNPs) on a whole genome scale. Specifically, we sequenced pooled genomic DNA using Illumina HiSeq2000 technology, aligned the reads against a reference transcriptome (also generated by NGS), and identified SNPs fixed and heterozygous in the pools of mutant and wild-type individuals, respectively. Given the high levels of synteny between lizard and snake genomes, we then used TBLASTX to search the *Anolis carolinensis* genome (the only squamate species with chromosome assembly) for orthologues of corn snake transcripts containing such SNPs. We confirmed these mapping results by genotyping 5 to 7 selected SNPs markers in all available offspring. This strategy further reduced the mapping interval for each mutation investigated and identified several promising candidate genes which were then analysed for differences in coding sequence and expression levels between wild-type and mutant individuals. Our study demonstrates that NGS is a powerful tool to identify genes of interest even in organisms with only limited genetic and genomic resources.

P-101 Mechanically gated ion channels during early Xenopus embryogenesis

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Cell shape changes and collective cell migrations are key mechanisms of the morphogenesis of tissues and organs. During animal development, these cell behaviors are highly coordinated and regulated. The central mechanism of these coordinations and regulations is interaction between cell adhesion and mechanical stress. It was shown that main sensors during mechanically activated cell migrations or extracellular matrix synthesis are mechanically gated ion channels. But practically all these data were obtain on *in vitro* models, and there are not evidences that these mechanisms could drive animal development *in vivo*. The aim of our work was to explore expression of mechanically gated ion channels during *Xenopus tropicalis* embryogenesis and identify, which of them could be involved in mechanically regulated morphogenesis. So far, we have checked the following ion channels: TRPV1, TRPV2, TRPV4, TRPM3, TRPM7, TRPC1-like, TRPC5, TRPC6, PKD2, Piezo1 and Piezo2. Maternally expressed following: TRPV1, TRPV4, TRPC6, TRPM7, Piezo1 and Piezo2. Relatively high expression level was detected for TRPV4, TRPM7 and Piezo1. TRPV2, TRPM3, TRPC1-like and PKD2 start express late: from neurula or hatching stage.We have shown that early morphogenesis could be regulated by mechanical stresses through mechanically gated ion channels and in future it should proved by gain and loss of function experiments.

P-102 Mechanisms of lateral organ laminarization in angiosperms: Zingiberales and beyond

Almeida, Ana Maria (UC Berkeley San Francisco, CA, USA)

Laminar expansion is a widespread phenomenon during lateral organ development in angiosperms. In leaves, for example, laminar expansion occurs as a result of balanced abaxial-adaxial gene expression. Here, we show the involvement of the abaxial-adaxial polarity gene network (GRN) in the evolution of stamen filament morphology in angiosperms. We also anticipate that co-option of the polarity gene network is a fundamental mechanism shaping many aspects of plant morphology during angiosperm evolution. Although the molecular mechanisms of laminar expansion have been largely uncovered, the resulting physical forces underlying such process are less known. Focusing on the downstream genes of the polarity GRN, we explored the potential physical processes resulting from this gene expression as well as the underlying dynamical patterning modules (DPM) associated with the physical process under study. We also discussed the relationship between DPM and GRN in plants and compared it to what have been described for animals. Finally, we performed a detailed analysis of the evolution of such DPM-related genes in angiosperms, and inferred correlations of the observed evolutionary pattern with the origin of new plant forms.

P-103 Minimal regulatory network predicts the differentiation and plasticity of T CD4+ lymphocytes

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T CD4+ lymphocytes orchestrate the immune response in mammals. They need to be capable of responding to changes in the environment by differentiating to give rise to different cell types depending on the immune response. They also exhibit plasticity in the response to changes in the signals of the microenvironment. We lack understanding of which are the key molecular elements of the underlying regulatory network and the mechanisms that underlie the differentiation and plasticity of these cells. We construct a minimal regulatory Boolean network consisting of transcription factors, signalling pathways, and cytokines whose steady states correspond to biological cell types of T CD4 cells. We provide a novel methodology to determine the key elements for the plasticity of the network, including important participation of SOCS proteins. Using this model we study how the cytokines present in the environment and perturbations of the elements of the network affect the transitions between steady states. Finally, we construct a cell fate map that captures part of the reported phenotypic plasticity. Development is not a static process; organisms develop in dynamic circumstances. Our model provides a theoretical approach to study the characteristics of the networks of complex biological functions in changing environments. A practical use of our results is the integrative approach for determining therapeutic targets for modulating the immune system.

P-104 Modulation of Platynereis larval behavior by neuropeptides

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Neuropeptides are neuronal signalling molecules present in almost all animal taxa. They are important modulators of many different behaviors and physiological processes, including sensorimotor integration, feeding and metabolism, central pattern generation, circadian rhythms and reproductive behaviors. Here I will study the neuropeptidergic modulation of behavior in larvae of the marine annelid *Platynereis dumerilii*. Platynereis belongs to one of the most understudied animal superphyla: the Lophotrochozoa. I focus on larval stages, which, despite their relatively simple nervous systems, exhibit a wide range of behaviors. All 99 neuropeptides found to date in Platynereis will be screened for their effect on behavioral responses to various environmental stimuli (e.g., light). Once the neuropeptides are broadly categorized according to their involvement in modulating responses to different stimuli (e.g., phototaxis), the nature of the response and the mechanism of neuropeptide action will be studied in more detail. These behavioral data will be combined with gene expression pattern and electron microscopy data to characterize the neural circuits modulated by neuropeptides. A global overview of neuropeptidergic behavioral modulation in a simple nervous system may provide insight into the role of neuropeptides in the evolution of early bilaterian behaviors.

P-105 Molecular and cellular differentiation during the early shell field development in *Lymnaea stagnalis*

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The evolutionary innovation of the molluscan shell supported the generation of a wide variety of molluscan life forms. However, its origins (both evolutionary and ontogenetic) are still poorly understood. Developmental specification of the larval shell-secreting organ (the shell field) appears to be evolutionarily conserved, often coinciding with an endoderm/dorsal ectoderm contact event following gastrulation that resembles a contact-mediated induction event. A deeper understanding of the gene regulatory networks that specify the shell field in a range of molluscan representatives would

provide deep insight into how the molluscan shell first arose. While several regulatory genes are known to be associated with larval shell formation (en, dpp, hox1), these are all expressed well after shell field specification. Preliminary data from the gastropod *Haliotis asinina* suggest an involvement of the contact-dependent intercellular signalling pathway Notch in shell field specification. Here, we examine the role of Notch signalling during shell field specification in the gastropod *Lymnaea stagnalis*. We show that cellular movements and differentiation events, which result in the specification of the shell field are likely to represent a contact-mediated induction event. Our preliminary data on the spatial expression of core components of the Notch pathway and its pharmaceutical inhibition do not support a role of Notch signalling in such a process for *L. stagnalis*. Rather, Notch signalling seems to function during early neurogenesis and cellular differentiation of the archenteron.

P-106 Morphological integration of sexual dimorphic traits in human skull

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The human skull is a complex integrated structure in which the existence of different modules has been postulated. Sexual dimorphism is unequally expressed in different craniofacial features. However, little is known about the degree of integration and modularity of dimorphic traits. Here, we employ a sample of 202 Spanish adult skulls of known sex that were analyzed using 2D geometric morphometric techniques (GM) in lateral view. A set of type I landmarks was recorded to describe two sexually dimorphic skull regions, the mastoid (MP) and zygomatic (ZP) processes, and then subjected to Generalized Procrustes Analysis (GPA). Integration and modular organization of both processes were studied between sexes using Partial Least Squares (PLS) analysis and permutational approaches based on the Escoufier RV coefficient. The strength of covariation between MP and ZP was computed using the Escoufier RV coefficient at different sample sizes (from 22 to 202 individuals). Results show that MP (RV=0.45; $p<0.001$) and ZP (RV=0.53; $p<0.01$) are integrated in the whole skull. Second, covariation is lower between ZP and MP (RV=0.17; $p<0.001$) than between each of these processes and the rest of the skull. Modularity tests suggest that these two processes could be independent modules, showing the minimum possible covariation. In addition, a clear pattern of decrease in RV coefficients at increasing sample sizes can be observed, showing a stabilization of RV values when sample size is larger than 100 individuals. Further, there are no significant sex-related differences in the degree of covariation between MP and ZP (IRVmale-RVfemalel=0.022; p=0.56) showing that the strength of covariation between modules is not different in both male and female traits. Based on the known sex Spanish skull collection, our findings show that different skull regions showing sexual dimorphism are distinct modules, the degree of modularity is the same in both sexes and the value of RV is stable at larger sample sizes.

P-107 Multi-level feedbacks during Tribolium segmentation

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Sequential segmentation occurs in vertebrates, arthropods and annelid worms, with conserved genes between these distant branches. A long-standing question has been how segmentation evolved, whether it arose in a common ancestor of the bilaterians and subsequently got lost in most animal lineages, or whether these three animal lineages developed a similar mode of segmentation in parallel. In this light, the short-germ insect Tribolium is an interesting subject for investigation. In contrast to Drosophila, where a hierarchical cascade of genes forms all (para)segments at approximately the same time, Tribolium forms its segments sequentially from a cellularized posterior growth zone, with clock-like expression of the segment-defining genes (Choe et al. 2006; El-Sherif et al. 2012; Sarrazin et al. 2012). Although the segmentation mode of Tribolium is reminiscent of that of vertebrates, there are some interesting differences: the involvement of pair-rule genes, the double-segment periodicity, and the near-simultaneous occurrence of tissue shape change (convergent extension) with segment formation. These differences may have important consequences for the segmentation process. In this project, we aim to elucidate the mechanisms of Tribolium segmentation by incorporating what is known from Tribolium into a multi-level computational model, and we fill in the gaps by drawing inspiration from what is known from vertebrates. Our tool is the Cellular Potts Model, which allows us to realistically model the interactions of cells in a tissue, and how properties of individual cells are determined by the dynamics of their genetic network. Using this framework, we recently showed how a segmented tissue pattern may drive convergent extension, demonstrating that the simultaneous occurrence of segmentation and morphogenesis leads to feedbacks between these two processes. We are currently further investigating feedbacks from convergent extension to segmentation. Our model results will not only tell us more about the properties of Trinolium segmentation, but may also shed light on the evolution of segmentation by highlighting both similarities and differences with vertebrate segmentation.

P-108 Multiple developmental roles of a tissue-specific alternative splicing factor across deuterostomes: ESRP genes are master regulators of diverse epithelial functions

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Alternative splicing (AS) greatly expands the proteomic and regulatory complexity of vertebrate genomes. A significant fraction of this AS is tightly regulated in a cell and tissue manner due to the action of a handful of tissue-specific RNA-binding proteins that regulate large networks of alternative exons. Despite their importance for embryonic development, and in contrast to what has been largely reported for transcription factors, previous comparisons of expression patterns of AS factors between distantly related groups have pointed to frequent acquisition of novel functions by these genes. Moreover, alternative spliced exons are highly evolvable and usually not conserved, even between mammalian species. Thus, understanding the evolution of the biological roles of AS factors and how their regulated target networks are assembled is a pending task in EvoDevo. The Epithelial Splicing Regulatory Protein (ESRP) gene family is expressed dynamically in diverse epithelia during mouse ontogeny. In cell culture experiments, ESRP1 has been shown to be a master regulator of Epithelial to Mesenchymal Transition (EMT), where it controls a network of alternative exons from genes involved in cell adhesion. We combine expression and functional data of ESRP genes in various deuterostome species. We show that ESRP expression is restricted to various types of epithelial cells in all studied species. However, at the functional level, these genes seem to be necessary for different developmental processes, involving epithelia, within the deuterostome clade. For example, in the zebrafish, ESRP2 knockdown causes pericardial edema. In Ciona, ESRP seems to negatively regulate Twist2, a transcription factor linked with cell migration in this species. In amphioxus, ESRP is expressed in the epidermic cells adjacent to the neural plate border during neurula stage. Finally, outside chordates, in sea urchin, we demonstrate that ESRP is involved in proper specification of the aboral ectoderm. In sum, our results exemplify how a master regulator with generally conserved tissue-type expression (i.e. epithelia) has evolved to regulate very diverse developmental programs during the evolution of the deuterostome clade.

P-109 Mushroom bodies: Homology or not?

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The mushroom bodies (MB) are prominent brain centers that can be found in various animals such as insects, annelids and onychophorans. Also in spiders a potentially homologous structure has been described. The reason behind the assumption of homology between different animal groups and naming those mushroom bodies was the description of this lobed neuropiles by Félix Dujardin in 1850 and the criteria for identification defined by Flögel in 1876. The question of homology has been controversial for some time. N ot all spiders have these specific brain centers and the function of the MB's differs significantly from the one known in insects like Drosophila. Therefore, no concrete answer has been found. In the spiders so far described with MB's, the connectivity is in the optic system. This is one of the reasons for some to challenge the idea of homology, as in most insects the MB's have a function in olfaction and olfactory learning. But new studies challenge this view as the variation of MB function within insects is large. During this study, we aim to show whether or not the MB's of the spider *Cuppenieus salei* are homologous to the MB's of other arthropods. With in situ hybridizations we examined and compared a number of genes with expression patterns described in Drosophila MB's during the embryonic development of *C. salei*. Our results show expression patterns for some of these genes during MB development in *C. salei* and might help to answer the question of homology the mushroom bodies.

P-110 Y-secretase activity is required for apical organ formation in the sea anemone *Nematostella vectensis*

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Apical organs are ancient sensory structures that are found in diverse animal phyla such as cnidarians, protostome molluscs and annelids, as well as deuterostome echinoderms and hemichordates. They are larvae-specific organs that are most likely involved in the detection of settlement cues and the induction of metamorphosis. Whether apical organs in different taxa share a common evolutionary origin is not clear and the molecular basis of their development is still poorly understood. We show here that γ-secretase is required for apical organ formation in the sea anemone *Nematostella vectensis*. γ-secretase is a multiprotein

complex responsible for the cleavage of transmembrane proteins within the cell membrane. Pharmacological inhibition of γ-secretase activity with DAPT causes a loss of the *Nematostella* apical organ and its ciliary tuft. Gene expression analysis shows that the cells expressing NvFGFa1, a key regulator of apical organ development, are not located to a coherent domain at the aboral pole, but are dispersed in a broad aboral territory upon DAPT treatment. Our findings suggest a model in which γ-secretase activity is required to maintain the integrity of the apical organ-forming domain, which provides a novel insight into apical organ development that might help to better understand the evolutionary history of these sense organs.

P-111 Next-generation approaches to understanding the evolution of germline

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Germline cells are unique as they can produce gametes and regenerate themselves, and can be specified by either maternally inherited determinants or by zygotic inductive signals. In the maternal inheritance mode, the germ cells are specified very early by the germ plasm synthesized during oogenesis. This mode is found in most model organisms (fruit fly, zebrafish, frog and nematode), whereas the zygotic induction mode may be the ancestral model of germline determination. Among the invertebrates, the only arthropod in which the germ line has been studied in detail is *Drosophila melanogaster*, which uses the maternal inheritance mode. However, this mechanism of germ cell specification is a derived feature in insects and seems to be limited Holometabola. The wasp *Nasonia*, like *Drosophila*, uses the maternal inheritance mode, and represents the most distantly branching holometabolous lineage relative to *Drosophila*. The assembly of *Nasonia's* germ plasm is dependent on a regulatory network that is very similar to that of *Drosophila*, and occurs in the same context of polytrophic ovaries, indicating that a regulatory network similar to both fly and wasp was present ancestrally in the Holometabola. Despite the overall similarity in *Nasonia* and *Drosophila* germline determination, some aspects are quite distinct, such as the morphology of the germ plasm, the formation of the pole cells, and the migration of the pole cells into the embryo interior. To characterization the ancestral and novel features of *Nasonia* germ plasm relative to *Drosophila* at the molecular the mRNA composition of the *Nasonia* oosome was characterized by RNAseq. The results confirmed that certain genes, such as oskar, nasos, etc., are conserved between the two species, and also revealed previously unknown genes, which could be crucial for the unique properties of *Nasonia* germ plasm and germline.

P-112 Nitric oxide-neuropeptide interaction in the settlement behavior regulation of the marine annelid, *Platynereis dumerilii*

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In marine invertebrate, neuropeptides and nitric oxide (NO) synthesized by the enzyme, nitric oxide synthase (NOS), act as critical internal regulators of settlement and metamorphosis. However, the coordinated interactions of these molecules are still poorly understood. To address this question, we are investigating the role of NO and its interaction with the settlement-inducing neuropeptide, myoinhibitory peptide (MIP), in the marine annelid, *Platynereis dumerilii* by integrating behavioral, cellular, and molecular biological analyses. Specifically, application of NOS inhibitors resulted in the repression of settlement behavior. In contrast, application of NO donor induced settlement behavior. Co-treatment of MIP and different concentrations of NOS inhibitors showed a concentration-gradient manner of settlement behavior repression. In situ hybridization analysis revealed NOS-expressing cells in the larval apical sensory organ (ASO), deeper inside of the ASO compared with MIP-expressing cells. These results suggest that NO is involved in induction of settlement behavior by being a downstream component of MIP. Ultrastructural analysis of NOS-expressing cell morphology, NOS knock-out line for in-depth behavioral analysis, and transcriptomic/proteomic analyses of gene/protein expression by NO and MIP exposures are currently underway.

P-113 NO chordate evolution

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Nitric Oxide (NO) is an ancestral and multifunctional signaling molecule involved in a lot of biological processes as development, neurotransmission (retrograde), metamorphosis, aging, regeneration, apoptosis and so on. In vertebrates, the biosynthesis of NO is catalyzed by three NOS enzymes with distinct functions and different territories of expression, although structurally highly similar: the constitutive endothelial eNOS and neuronal nNOS, regulated by intracellular Ca+ increase, and the inducible iNOS, which is activated under stressful conditions. These proteins are highly conserved in Metazoan both structurally (functional domain organization, number and phase of introns) and functionally. To better understand the evolution and function of these genes in chordates, we choose a "living fossil" model organism as the cephalochordate Amphioxus that possess two neuronal NOS-A and NOS-C genes, and one inducible NOS-B, that

are probably products of a specific-cephalochordate gene duplication. The canonical vertebrate endothelial NOS seems then to be either lost in amphioxus or, most parsimoniously, is a vertebrate's invention appeared in parallel with a more complex circulatory system. We are in progress to characterize the NOSs developmental expression profile using embryos from the mediterranean Amphioxus species - *Branchiostoma lanceolatum* - by Q-PCR and in situ hybridization experiments. The NOS-C contains a PDZ domain that is typical of the neuronal vertebrate NOS, and start to be expressed at neurula stage (20 hpf) at the level of the anterior part of the neural plate, the neuropore (NP). Later in development at pre-mouth stages (48 hpf), the signal is present in the cerebral vesicle and extends posteriorly along the nerve cord up to the first pigment spot (PS). In the larval stages, at 3 days post fertilization, the expression in the nervous system become weaker while it increase at level of club-shape gland (CG). The expression pattern of the other two genes, NOS-A and NOS-B will be investigated as well. On the other side we are trying to interfere with the endogenous activity of the Nitric Oxide using different drugs and preliminary results show that *in vivo* deprivation of NO is involved in the development of the mouth at 2-3 days post fertilization. To this aim we treated amphioxus embryos with several drugs like L-NA, L-NAME, TRIM, SPERNO in order to decrease or increase the endogenous NO level, or LPS to simulate a strong bacterial attack. The phenotype will be analyzed using morphological techniques as Scanning Electron Microscopy (SEM) or molecular approaches by specific tissue-specific markers (SoxB1 and Hh).

P-114 Nodal signaling regulates the innate asymmetry of the Amphioxus pharynx

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In vertebrates, Nodal signaling regulates left-right axis specification via activation of left-sided expression of Pitx transcription factors. This pathway is then responsible for left-right asymmetry and left-sided positioning of internal organs. The basal chordate amphioxus develops asymmetrically arranged pharynx with left-sided oral opening and preoral pit, and right-sided endostyle and club-shaped gland. To elucidate whether Nodal-Pitx pathway plays role in establishment of the asymmetric pharynx in amphioxus, we treated early neurulae with Nodal inhibitor SB505124. This treatment resulted in symmetrically developing pharynx and a loss of left-sided oral opening and preoral pit. Concomitantly, expression of pharyngeal left-sided markers (Pitx, Dkk1, Lhx3) was lost and, instead, the left side expressed right-sided markers (Krox, FoxE4, FoxQ1). This apparent duplication of the right side implies that Nodal is necessary for the induction of the left side and thus establishes the innate asymmetry of the amphioxus pharynx. Apart from the necessity, we are currently testing whether Nodal signaling is sufficient to induce the left pharyngeal side to get more complete picture of the left-right axis induction at the base of chordate phylogeny.

P-115 Non-canonical dorsoventral patterning in the moth midge *Clogmia albipunctata*

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Étienne Geoffroy Saint-Hilaire famously proposed that invertebrates and vertebrates had very similar body patterns, but with an inversion in the dorsoventral (DV) axis. This idea is nowadays supported by molecular evidence showing that key factors for DV patterning (such as BMP morphogens) are expressed with opposite polarity in these two groups of animals. We have recently reported surprising evidence that the situation may not be quite as clear-cut. The insect BMP homologue dpp is expressed ventrally, around the anterior and posterior poles, in the blastoderm of the nematoceran moth midge *Clogmia albipunctata*. How this arrangement of gene expression in *Clogmia* is able to function as a dorsal morphogen gradient is unknown. We are currently systematically characterising other components of the DV patterning pathway to elucidate the mechanism of Dpp gradient formation in this fly. Our analysis suggests that a shuttling mechanism like that proposed for Drosophila is improbable in *Clogmia*. We are using RNAi, previously unavailable in this species, to elucidate the interactions between these genes and improve our understanding of the variety of patterning mechanisms available to specify the DV axis.

P-116 Non-invasive long-term fluorescence live imaging of *Tribolium castaneum* embryos

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Insect development has contributed significantly to our understanding of metazoan development. However, most information has been obtained by analyzing a single species, the fruit fly *Drosophila melanogaster*. Embryonic development of the red flour beetle Tribolium castaneum differs fundamentally from that of *Drosophila* in aspects such as short-germ development, embryonic leg development, extensive extra-embryonic membrane formation and non-involuted head development. Although *Tribolium* has become the second most important insect model organism, previous live imaging attempts have addressed only specific questions and no long-term live imaging data

of *Tribolium* embryogenesis have been available. By combining light sheet-based fluorescence microscopy with a novel mounting method, we achieved complete, continuous and non-invasive fluorescence live imaging of *Tribolium* embryogenesis at high spatiotemporal resolution. The embryos survived the 2-day or longer imaging process, developed into adults and produced fertile progeny. Our data document all morphogenetic processes from the rearrangement of the uniform blastoderm to the onset of regular muscular movement in the same embryo and in four orientations, contributing significantly to the understanding of *Tribolium* development. Furthermore, we created a comprehensive chronological table of *Tribolium* embryogenesis, integrating most previous work and providing a reference for future studies. Based on our observations, we provide evidence that serosa window closure and serosa opening, although deferred by more than 1 day, are linked. All our long-term imaging datasets are available as a resource for the community. *Tribolium* is only the second insect species, after *Drosophila,* for which non-invasive long-term fluorescence live imaging has been achieved.

P-117 Novel beta-glucosidases of the family GH3 could be involved in the development of various animal groups

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Glycoside hydrolases (EC 3.2.1.-) are enzymes which catalyse the hydrolysis of the glycosidic bond between two carbohydrates or between a carbohydrate and a non-carbohydrate moiety. Based on amino acid sequence similarities currently they are classified into 133 protein families in Carbohydrate-Active enZYmes (CAZy; http://www.cazy.org/) database. Proteins from the family GH3 are important bacterial, fungal and plant enzymes involved in cell wall remodeling, energy metabolism and pathogen defense. The core of GH3 enzymes is formed by two subunits, both participating on the catalytic center formation. Domain1 is typical (α/β)8 barrel and domain2 adopts a five-stranded α/β sandwich fold. Various additional C-terminal domains, including evolutionary conserved fibronectin type III (FnIII) domain, contribute to structural variability of GH3 enzymes. In plants they form multi-gene families comprising more than 15 genes in monocot and dicots species and more than 6 genes in lower plants. Although almost 6,000 GH3 proteins were identified and more than 200 of them have been characterized so far, until now they have been unknown in animals (Metazoa). Using bioinformatics approach we found many novel GH3 proteins in various animal groups from Porifera to some vertebrates, predominantly birds and reptiles. Intriguingly, they are missing in such important groups like arthropodes and mammals. This patchy distribution could be either result of extensive gene loss or horizontal gene transfer. Based on our phylogenetic analysis as well as study of domain structure, conserved sequence regions composition and tertiary structure comparison we assume that the animal GH3 proteins are most likely beta-glucosidases with C-terminal fibronectin type III (FnIII) domain. Although their biological function is still unknown, transcriptomics data show gene expression in early stages of embryogenesis. It is thus possible that at least some of these genes could participate in animal development.

P-118 Novel mechanism of TCF function in *Ciona intestinalis*

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We have previously observed TCF/ β-catenin mediated repression in the ascidian *Ciona intestinalis* (Rothbächer et al. 2007). We are now studying the molecular basis of the largely unknown process of "opposite" gene transcriptional regulation by TCF, using the developing embryonic ectoderm of the developing *C. intestinalis* as a model system. Ascidians are tunicates and the closest sister group to the vertebrates within chordates sharing a common larval body plan with these (including a dorsal neural tube and notochord). Their fixed embryonic lineage with few and large blastomeres allows for a cellular resolution of molecular mechanisms during embryonic development. The compact genome with good access to regulatory regions is fostering the analysis at transcriptional and translational level of such novel aspects of TCF function. "Classical" TCF target genes are activated by the canonical Wnt pathway through TCF/ β-catenin binding at known TCF binding sites and repressed by TCF in the absence of Wnt (and β-catenin). Another set of direct TCF targets has recently been observed that displays "opposite" regulation, that is repression by TCF/ β-catenin in presence of Wnt, (and possibly activation by TCF in absence of Wnt). First hypotheses are proposed in worms. Interestingly, canonical TCF sites are not involved in these cases. We are now trying to identify and describe the molecular mechanisms of TCF mediated "opposite regulation" of gene transcription in the chordate context, during ectoderm and nervous system formation in *C. intestinalis*.

P-119 Novel mutation in human AMELX gene is associated with defect in amelogenesis

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Amelogenins comprise closely related proteins involved in tooth development, specifically in amelogenesis, the development of enamel.

Among eutherians, amelogenins are well conserved. The amelogenin genes (AMEL) are studied in mammals as important markers of sex chromosome evolution, in the context of the pseudoautosomal boundary and differentiation of X and Y chromosomes. The AMEL genes, AMELX and AMELY have been most widely analyzed in humans. The X chromosome form (AMELX) is located at Xp22.1-Xp22.3. Up to now, a number of sequence variants at AMELX are described, some of them associated with amelogenesis imperfecta, a hereditary disorder characterized with disturbed enamel formation. However, more information on specific genotype/phenotype correlations is still needed. In this study, we have analyzed DNA samples of a family with disorder in enamel development. Direct sequencing revealed the presence of the novel mutation, the substitution T>C in third intron, at position c.103-3. This mutation is located close to the splicing site and *in silico* prediction shows 0.90 probability that it is disease causing. Our data show that variation in noncoding AMELX sequence could have signifficant effect on the development of enamel. Further functional analyses are required to elucidate the specific correlations between genotype and phenotype.

P-120 Origins and regulation of an eutherian novelty: The BGW cluster

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Two related gene subfamilies known as BEX and TCEAL (also known as WEX) map to a genomic region specific to Eutheria (placental mammals), located on the X chromosome. These families are part of a gene cluster named "BGW cluster", together with the ARMCX family and HNRNPH2. Some of the BEX/TCEAL genes have been related to control the balance between proliferation and differentiation while others promote apoptosis in a p75-dependent manner, but most of them remain poorly studied. The ARMCX family and HNRNPH2 are derived from retrocopies of the ARMC10 and HNRNPH1 genes respectively — conserved across bilateria, and located in autosomal chromosomes — whereas no orthologs have been found for the BEX/TCEAL family outside of Eutheria. However, all these genes share an intriguing feature: a sequence motif in their proximal promoter region that appears to be crucial for their expression, the BGW motif. To further understand the evolution of this gene cluster, we investigated the origin of the BEX/TCEAL genes and traced it to an atypical formation in the ancestor of eutherians. Furthermore, novel features associated with BEX/TCEAL suggest a more complete scenario for the origin of the cluster: the BGW motif was already present at the HNRNPH2 locus in the ancestor of therian mammals, being subsequently duplicated and coopted in the eutherian lineage by the BEX/TCEAL ancestor and, posteriorly, by the ARMCX ancestral gene. Finally, we also studied the expression of the BEX/TCEAL genes during mouse development using in situ hybridization. We found that they are highly expressed in the brain and placenta, which are structures that require a well-tuned control of cell cycle during their development in eutherian mammals. Here we propose a scenario for the origin of the BEX/TCEAL family and for the formation of the BGW cluster where they belong. Their uncommon origin, their pattern of expression, and their putative biological function during development makes these genes an interesting subject of study to understand how lineage-specific genes could contribute to mammalian evolution.

P-121 Origins of modularity in the Soay sheep skull

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The semi-autonomy of the parts, or modularity, is essential for the diversification of phenotypes between and within taxa. Two types of modularity will be investigated in this study: developmental and functional. Developmental modularity pertains modules that share embryological origins or signalling between tissues, whereas functional modules execute similar functions, or are under similar mechanical loads. Some of the main questions addressed in modularity studies are: (1) whether functional modularity is related to developmental modularity (2) and whether the modules explain morphological variation in the clade. The modularity hypotheses proposed here divide the skull in face and neurocranium (developmental modules), since their cells originate from neural crest and paraxial mesoderm respectively; and oral region, upper face and braincase (functional modules). The aims of this study are first, to test which hypothesis is true for either/both functional and developmental modules in Soay sheep's skull; and second whether in either of these modularity types there are differences between sexes and horn morphs. The results show that the skull of Soay sheep is divided in face and neurocranium modules, in both functional and developmental components.
Both sexes and all horn morphs in both types of modularity fit the „developmental" hypothesis. Because developmental processes form the structures that perform functions, developmental modularity is expected to influence functional morphology. The division of the skull in face and neurocranium in wolves is apparently linked to the great morphological variation seen in dog breeds, and the presence of the same modules in sheep could also explain the vast variation on the skull among sheep breeds. Earlier results in primates, rodents and carnivores suggest that these patterns of morphological integration may hold in many other mammalian species based on the similarity of craniofacial growth.

P-122 Osteogenic differentiation of oral mucosal mesenchymal progenitor cells

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Objectives: To explore the osteogenic differentiation ability of mesenchymal progenitor cells *in vitro* and *in vivo*, which were derived from oral mucosal lamina propria.
Methods: Isolate mesenchymal progenitor cells from rat oral mucosal lamina propria (OMLPPC). Subsequently, OMLPPCs were subcultured to third-passage; some were implanted onto scaffolds, and then implanted into dorsal subcutaneous area of athymic mice. Some were cultured in osteogenic inductive media, and analyzed by morphology, immunohistochemistry and alizarin red staining methods.
Results: After osteoblast-inducing culture, OMLPPC became larger and multiangular. Induced for 7 days, the type I collagen appeared positive by immunocytochemistry. The mineralized nodules were verified by alizarin red after 21 days. After 8 weeks implanted *in vivo*, the OMLPPCs with scaffolds exhibited osteogenic differentiation, and formed bone-like structures.
Conclusions: OMLPPC have the potential of osteoblastic differentiation *in vitro* and *in vivo*. OMLPPC may serve as a suitable source of cells for future bone tissue engineering strategies.

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P-123 Overexpression of human scute homolog genes in Drosophila

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Even when humans lose most of their fur to become the naked ape, they still keep human body hairs, such as axillary hair, pubic hair and beard. It is unclear which selector genes target the sites for the hair growth. Although the genetic differences in the human are not yet known, we do know that Drosophila uses a single genetic locus — the Achaete-Scute Complex, to demarcate the territories where bristles can form throughout the body. It is presumed that comparable centers exist in human genome. By using bioinformatics searches, we find that there are five isoforms of Drosophila Achaete-Scute homologs in human. From a phylogenetic tree, we recognize that hASH1 and hASH2 come from the same ancestral gene, while hASH3-5 originates from a different ancestral gene. To test whether one of these isoforms might be responsible for hair growth, we cloned the human Achaete-Scute homologs (hASH1) and hASH4 into a pUASg-HA.attB vector and made transgenic flies containing hASH1 and hASH4 respectively. When combined with the pnr-Gal4 driver, which gives an expression pattern in the notum, we find that the antibody staining in both transgenic lines show the expected expression pattern in lower part of the wing disc identical to expression of the gene pnr. In adults, we find that overexpression of hASH1 induces additional macrochaetes on the fly notum, which is similar to the patterns after overexpression of the Drosophila scute gene. However, overexpression of hASH4 did not produce extra macrochaetes on the notum of flies. By comparing protein structures, we see that both human Achaete-Scute homologs have the basic helix-loop-helix (bHLH) domains but hASH4 has a shorter sequence in c-terminal motif, implying a possible loss of proneural function in hASH4 due to the lack of a c-terminal motif. Our results suggest that, due to duplication of this proneural gene, the isoform of hASH1 has kept the function in hair (bristle) growth. The isoform of hASH4 has evolved to fulfill a divergent function, which is no long essential for hair (bristle) growth and mainly contributes to regulation of skin development.

P-124 Phenotypic divergence among spadefoot toad species reflects accommodation of mechanisms underlying developmental plasticity

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Selection in heterogeneous environments favours plasticity as it allows organisms to adapt to rapidly changing conditions. Developmental plasticity allows populations to withstand rapid environmental changes and confers an overall faster rate of adaptation. Conversely, if plasticity costs are high and the environment stabilises, selection results in genetic assimilation, which could result in trait divergence and species diversification. Current evolutionary theory contemplates that phenotypic divergence between species may initiate as environmentally-induced expression of alternative phenotypes. Descendant lineages of a plastic ancestor evolving in stable divergent environments may lose plasticity over time, their development becoming speciali s ed to produce fixed phenotypes matching each environment. In that case, we would expect ancestral plasticity to mirror differences among taxa and that the same mechanism allowing ancestral plasticity was also the main mechanism explainig species divergences. In that light, we are study ing mechanisms of plasticity behind the evolutionary divergence of spadefoot toads. Old World species (*Pelobates*) breed in long lasting ponds and have long but plastic larval periods, whereas New World species (*Scaphiopus*) have

specialised in ephemeral ponds and have evolved very short larval periods. We hypothesise that *Scaphiopus* has undergone genetic accommodation of ancestral plasticity, which has resulted in canalised short larval periods. To test this hypothesis we have studied the mechanisms underlying developmental acceleration in response to pond drying and compared it across species. We have found that *Pelobates* tadpoles, which reflect the ancestral state of the group, increase their metabolic rate, and thyroid hormone and corticosterone concentrations in response to decreased water levels. All these parameters, however, seem to have been canalised in *Scaphiopus*, lending support to the hypothesis of genetic accommodation.

P-125 Phenotypic divergence is triggered by a bidirectional parental dominance in the transcriptomes of sibling orchid allopolyploids

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Hybridization and polyploidization have been central to the evolution of angiosperms, starting with their origin. Immediately following a polyploidization and/or a hybridization event, a genome suffers adjustments in organization and function, thereby influencing the adaptive success and the evolutionary fate of resulting lineages. Most allopolyploids have multiple origins, but the long-term significance of iterative allopolyploid evolution is not fully understood. We comparatively investigate here gene expression alterations in morphologically- and ecologically-divergent, sibling allopolyploids *Dactylorhiza majalis* and *D. traunsteineri* (Orchidaceae), together with representatives of their diploid parents, aiming to understand their importance to the phenotypic divergence of the polyploids. For this aim we use a high-throughput, NGS based, RNAseq experiment, including 29 individuals collected on a European-wide scale, after they have been grown in uniform conditions for a full growing season. Our extensive RNAseq experiment read in total close to 10 billion 100 bp Illumina HiSeq reads. The gene expression data from each individual were mapped against a de novo assembled diploid transcriptome reference, and gene expression has been quantified using the CLC Genomics Workbench. Differential gene expression was estimated by fitting their variance on a negative binomial distribution using DESeq and edgeR in the Bioconductor. We observe a trend of increased overexpression of genes in the younger polyploid *D. traunsteineri* in comparison to *D. majalis*, whose transcriptome generally resembles more closely those of the diploid parents. The phenotypic divergence between the polyploids seems mediated by a general parental dominance in opposite directions in the sister polyploids, a pattern retained partly also at the level of transgressively expressed genes. Significantly upregulated genes in *D. traunsteineri* as compared to *D. majalis* include some genes related to molecule binding, catalytic activity and transporter binding, some of potential ecological relevance. The massive expression differences among the diploid parents became reconciled in the sibling *Dactylorhiza* polyploids in different ways, thereby producing an array of different ecological and morphological properties which can set the stage for further differentiation, most probably in response to selection and/or drift. Overall, our data brings evidence that recurrent allopolyploidizations can modify transcript architecture in sibling allopolyploids in different ways, progressing according to the evolutionary age of the polyploids.

P-126 Photoreceptor cell evolution in ambulacrarian larvae, a first contribution

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An intriguing biological topic is the origin and evolution of sensory systems. Opsins are key proteins mediating the detection of light among Eumetazoa. In this study, we focus on the evolution of interactions between putative photoreceptor cells and the nervous system within Ambulacraria, a deuterostome clade where the larval and juvenile photoreceptor apparatuses might have arisen independently. In the sea urchin *Strongylocentrotus purpuratus*, previous studies have demonstrated that 2 out of 6 *bona fide* opsin genes, the ciliary SpOpsin1 and the rhabdomeric SpOpsin4, are expressed in juvenile and adult tube feet, pedicellaria and spine base associated with the skeleton. Here we show that 4-arm pluteus larvae of *S. purpuratus* possess two symmetrical SpOpsin1-positive cells within a conspicious aggregation of nerve cells, the so-called lateral arm cell clusters. Moreover, we present a yet partial, although quite comprehensive phylogeny of ambulacrarian opsins aimed at assessing orthologies, with the final goal of identifying gene novelties and/or modifications allowing these animals to adapt to new habitats and lifestyles. Future studies will address the identification of opsin-expressing cells of these ambulacrarians at different developmental larval stages. Sp-Opsin knockout larvae will allow us to investigate correlations between photoreceptors and organismal behaviours (e.g., ciliary beating and diel vertical migration). The combination of these multidisciplinary approaches will allow us to shed light on deuterostome photoreceptor cell evolution, and identify potential homologies among the dipleurula larvae of these groups.

P-127 *Pipistrellus pipistrellus* (Chiroptera, Vespertilionidae) postnatal baculum development

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The baculum (os penis) is a heterotopic bone in the glans penis, which is probably subject to sexual selection and may play a role in reproductive isolation between closely related species. The cryptic species complex *Pipistrellus pipistrellus* is a remarkable model system for investigating links between morphology and niche partitioning. Studying baculum development may also complement research on the mechanical function of this bone. In this ongoing study we will compare baculum histomorphology of juvenile and sub-adult *P. pipistrellus* to that of adults bats, and to *P. nathusii*, *Myotis myotis*, and *M. emarginatus*. Correlative imaging, validating quantitative microCT images with undecalcified, surface-stained, serial ground section histomorphology, of our preliminary sample of *P. pipistrellus* (adult n=65, sub-adult n=8, juv. n=2), has yielded the following results. In *P. pipistrellus*, the whole baculum of juvenile and sub-adult bats consists of woven bone. While the distal part of the shaft with the forked tip seems to be mostly developed, the proximal base of the baculum is distinctly different from the adult appearance. The branches are shorter and less broad than in the adult specimens. The base consists of densely packed, large, round osteocytes. In some of the bacula, a small medullary cavity was found where the branches of the base meet the shaft, sometimes with a large opening to the ventral side of the bone. Some bacula did not have a medullary cavity at all. Our preliminary results on baculum development show that the distal part of the baculum reaches its adult shape before the proximal part. The different states of medullary cavity development we found suggest the medullary cavity first forms from the ventral side of the baculum, where the branches of the base meet the shaft. Our sample did not include young enough individuals to confirm if a cartilage precursor to the baculum exists in this species. Most of the sub-adult bats we studied were already capable of flight and had probably already left the nursery colonies in which they were born.

P-128 Placode size evolution in *Astyanax mexicanus* blind cavefish

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The fish *Astyanax mexicanus* presents, within the same species, several populations of river-dwelling surface fish and blind cave-living fish. In blind cavefish, the eyes first develop almost normally during embryogenesis. But 40 hours after fertilization (hpf), after the embryo has hatched, the lens enters apoptosis, which triggers the progressive degeneration of the entire eye. The mechanism leading to lens apoptosis is unknown, but should take place during the early stages of lens development. The lens develops from a placode, a thickening of the ectoderm at the neurula stage. All placodes, giving rise to sense organs of the head, originate from the "panplacodal" field, located at the border of the anterior neural plate at 10hpf. We compared the patterning of the panplacodal field in the 2 morphs, using in situ hybridizations for placodal marker genes. In cavefish, the lens placode territory is reduced at 10 hpf and the lens is smaller at all stages examined. Conversely, the olfactory placode is enlarged and gives rise to a bigger olfactory epithelium in the cavefish larva. We reasoned that modifications in placode size in cavefish arise very early, at the end of gastrulation. We focused on 3 signaling molecules (Shh, Fgf8, Bmp4) with spatio-temporal expression differences between cave and surface embryos. Pharmacological treatments with inhibitors of these signaling pathways show that Shh and Fgf signaling are responsible for placode size variation in cavefish. In sum, our data show that quantitative variations in the relative importance of sensory systems are due to small and subtle differences in signaling systems during early embryonic development.

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P-129 Positive selection drives gene duplications to fixation in Drosophila

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New genes are a major source of genetic innovation and are important contributors to evolutionary novelties. New genes play key roles in ecological adaptations, in the evolution of developmental innovations and of new morphological structures. Despite their clear evolutionary importance, there are fundamental gaps in our understanding of how new genes are created and how they become fixed in populations. These gaps stem from the difficulty in studying new genes while still in their polymorphic stage, and therefore still bearing the hallmarks of the mutation and selection processes that shaped their early evolution. Because most new genes are created by duplications and deletions, we set out to identify copy number variants in the genomes of 84 *Drosophila melanogaster* lines originating from five populations from five different continents. Among the ~60,000 variants that we identified (and extensively validated), there are ~500 polymorphic new genes. Most new genes correspond to complete gene duplications,

but we have also identified retrogenes (duplications with an mRNA intermediate) and chimeric genes created by the juxtaposition of the coding sequence of two parental genes. By combining different population genetic approaches we infer that positive selection, and not genetic drift as posited by most models, drives new gene duplications to fixation. Using gene expression arrays, we investigate the role played by changes in dosage in the fixation of new gene duplications.

P-130 Pro-differentiation state of Deuterostomes´ nervous system in an evolutionary perspective

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Comparative genomic analysis in Deuterostomes shows several conserved regulatory regions close to important developmental genes. Nevertheless, the regulatory mechanisms that orchestrate developmental genes remain still not fully understood. In 2011 it was recognized for the first time the highly conserved regulatory region of SoxB2, an important gene involved in early specification of the nervous system (Royo et al., 2011). Based upon the fact that the zebrafish's developmental machinery was able to recognize conserved non-coding regions (CNR) from evolutionary distant animals as sea urchin, amphioxus and human in transgenic experiments, we made an attempt to recognize the transcription factors (TFs) that bind to those regulatory fragments in the ascidian Ciona intestinalis and in the sea urchin *Strongylocentrotus purpuratus*. It is remarkable that even if the *Ciona* genome is missing the conserved SoxB2-CNR, transgenesis experiments in this species revealed that the amphioxus' SoxB2-CNR is able to drive the expression of a reporter gene in putative neuronal cells. To characterize its regulation we performed a deep bioinformatic analysis searching for TFs putative binding sites. We identified sequence fragments that could be the active core of the SoxB2-CNR, and we are in the process of testing their regulatory activity *in vivo*. In sea urchins, we characterized the SoxB2 expression pattern at different developmental stages by in situ hybridisation. We knocked-down SpSoxB2 by injection of morpholino antisense oligonucleotides (MO) in order to characterize the morphants phenotype using pan-neural markers. Our goal is to demonstrate the conservation of the SoxB2 regulatory network among Deuterostomes. On the other hand, we cannot exclude that different results will show up, as a set of regulatory factors that could have changed in different species, altered during the 500 MY of evolution, despite the structural and expression pattern similarity of SoxB2.

P-131 Quantitative mechanisms explain system drift in the dipteran gap gene network

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Dipterans (flies, midges, and mosquitoes) determine segmentation along their main body axis during the blastoderm stage of embryogenesis. In the initial phase of segmentation maternal gradients are interpreted by the gap genes, the first set of zygotic genes that divide the embryo in antero-posterior regions. Analysis of spatio-temporal expression and systematic knockdown of these gap genes in the scuttle fly *Megaselia abdita* reveals quantitative differences in gap gene cross-regulation between this species and *Drosophila melanogaster*. We have used mathematical modelling and reverse engineering of this gene network to gain insight into the regulatory mechanisms that underlie the observed differences in expression dyamics between the two species. Our models confirm that the qualitative structure of the gap gene network is conserved. Differences in expression dynamics are fully explained by distinct initial placement of the gap gene expression domains, and quantitative modifications of specific interactions. Our models agree with the available evidence from RNAi, and go further as they provide a precise understanding of the causal effects of each change in interaction strength. This is the first time that anybody has been able to study the mechanistic effects of such subtle changes at the level of an entire evolving developmental gene regulatory network. In addition, it provides a new perspective on regulatory evolution by system drift, where instead of changing the nature of regulatory interactions, the system is constrained to changes in the ratios of interaction strengths.

P-132 Rearing pups with fostering mothers reveals novel parent-of-origin effects in early vocalizations and attachment behavior

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Genetic variations are responsible for various behavioral differences in early development and adulthood. Recently, epigenetic mechanisms have also received particular attention for their role in modulating behavior. In particular, over the last few years we have investigated epigenetic parent-of-origin effects on behavioral and physiological traits and discovered dramatic effects of parental genetic background. Here we studied whether rearing pups with biological versus fostering mothers has effects on parent-of-origin traits. C57Bl/6N and Balb/C

are two classical strains known to differ mainly for maternal care and anxiety. C57Bl/6N show high level of maternal care and low anxiety, while BALB/c exhibit low level of maternal care and high anxiety. We studied C57Bl/6N and BALB/c mice and their reciprocal crosses reared with biological mothers or with CD1 fostering mothers. We have investigated early ultrasonic vocalizations (USVs), attachment behavior in a novel Mouse Attachment Test (MAT) and the anxiety profile in a classical Open Field (OF) test. We have found that fostering reduced the amount of early postnatal calls in both reciprocal crosses. Fostering induced also a reduction of anxiety later after birth and, interestingly, this was maternally driven. Furthermore, in the MAT, fostered mice showed an increase of the exploration of a stranger female, with whom it was left for three minutes. When the pups were reunited with the mother, the time spent with her varied according to genetic and parental origin if the pups were reared with CD1 foster mothers; furthermore fostering reversed some genetic differences in the preference for the mother rather than the stranger. Anxiety levels measured in the OF test revealed a similar trend. Classical fostering and cross-fostering experiments suggest that the adult phenotype is dictated by a rearing mother-driven phenotype. Here, for the first time, we show a different effect of fostering. In particular, fostering experiments combined with reciprocal crosses strategies highlight specific parent-of-origin genetic differences in early behaviors and different measure of attachment and anxiety.

P-133 Receptor mediated endocytosis of Wnt

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The patch-clamp capacitance technique is an important tool to analyze the molecular mechanisms of exo- and endocytosis. This technique takes advantage of the fact that exo- and endocytosis are associated with changes in plasma membrane area leading to proportional changes of electrical membrane capacitance. In high-resolution microscopic capacitance measurements patch-clamp capacitance measurements can even be used to resolve the fusion and fission of single vesicles in real time. We use capacitance measurements in to analyse receptor-mediated endocytosis of Wnt ligands. Our analysis revealed that different Wnt ligands induce a strong and fast response in patch-clamp capacitance measurements. These results could be confirmed using confocal imaging with endocytic markers. We used macroscopic and microscopic capacitance measurements to monitor the endo- and exocytotic activity upon Wnt stimulation. In macroscopic whole-cell recordings we could show that the membrane capacitance increases immediately after adding the Wnt proteins. In microscopic cell-attached measurements the endocytosis was extremely up regulated. In these high-resolution capacitance measurements not only the frequency of events can be analysed but also the size and kinetic of the vesicles is detected. It is also possible to discriminate between transient forms of exo- and endocytosis, also called "kiss-and-run", and permanent fission and fusion events. Upon Wnt-stimulation many permanent endocytic vesicles were visible and most of the events occurred within a very short time frame.

P-134 Regulation of internode morphogenesis in colonial hydroid *Dynamena pumila* L. (Hydrozoa, Cnidaria)

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There are many hypothesis proposed explaining mechanisms of morphogenesis regulation in different organisms during their development or regeneration. Among them two main groups can be distinguished. The first one rests on the assumption of morphogen gradients. The second group considers the morphogenesis as self-organising processes. The hypothesis proposed by Beloussov (Beloussov et al., 1993, 2003), belonging to the second group, asserts that morphogenesis in hydroids (which is based on growth pulsation) is regulated by the mechanical interactions within the rudiment: contraction/extension forces in epithelial layers, changes in the geometry of the rudiment, development in the direction of mechanically stable states. In the present work we experimentally tested the hypothesis of Beloussov, that the partitioning of the shoot apex into three rudiments in the colonial hydroid *Dynamena pumila* is interconnected with increasing of its size. We experimentally prevented the apex size increasing expecting that the morphogenetic cycle will end by subdivision of the apex into fewer rudiments. Our results testify against the idea that the mechanical interactions solely regulate internode morphogenesis in *D. pumila*. There is a certain morphogenetic programme independent from physical parameters of the tissue.

P-135 Retention of ancestral developmental potential for dentition in the teleost fish (*Astyanax mexicanus*)

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Dentition in ray-finned fishes was ancestrally widespread throughout the oropharyngeal cavity, with the predominant evolutionary trend being tooth loss in the central region and retention in anterior and posterior ones. Reversal of this trend is rare but has occurred in a number of groups. Previously we showed that competence to respond to transgenic overexpression of a tooth initiation signal (the TNF family ligand Ectodysplasin - Eda) with the production of ectopic

teeth is limited to the posterior pharynx of the zebrafish. This result is consistent with the evolutionary conservation of tooth location in the order Cypriniformes, to which the zebrafish belongs. Here we show that similar overexpression of Eda in the Mexican Tetra (*Astyanax mexicanus*), a member of the related order Characiformes, results in appearance of ectopic teeth in the central oropharynx, both on anterior gill arches and several bones of the palate. Among these latter bones are ones that variably bear teeth in characiforms as well as others on which teeth have been regained after long absence in some advanced spiny-rayed fishes. Our results suggest variable retention of the developmental potential for dentition among lineages of fishes. In addition, they implicate alterations in Eda signaling in the loss and reappearance of teeth in vertebrate evolution.

P-136 Revisiting HOX cluster evolution in Nematoda

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HOX genes are homeodomain transcription factors that are involved in the specification of anterior-posterior patterning along the body axis during early embryogenesis in bilateral animals. They are usually clustered on the genome and the order of genes in the cluster is conserved in most phyla. In contrast, in *Caenorhabditis elegans* (phylum Nematoda) the HOX genes are not tightly clustered, and five orthologue groups are absent. Previously, PCR-based surveys of diverse nematodes have shown that several of these losses occurred piecemeal within the phylum. The emergence of high-throughput sequencing technologies during the last decade has resulted in the determination of draft genome sequences for a large, diverse sample of nematode species. Here, using these genome sequences, we present a comprehensive survey and phylogenetic analysis of HOX genes from all major clades of the Nematoda, and propose a hypothesis for the evolutionary trajectory of this gene cluster. These findings might serve as a resource for understanding the underpinning mechanics of gradual gene loss within highly conserved gene families, and the evolution of the atypical mode of development in Nematoda.

P-137 Role of adaptors Shc, Dos and Drk in Torso and EGFR signaling in Tribolium

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In the genome-wide RNAi screen iBeetle (http://ibeetle.uni-goettingen.de) we found that knockdown of Tc-Shc („SH2 domain containing protein") gave rise to embryos with segmentation defects very similar to those of Torso, i.e. lacking all posterior/abdominal structures and some thoracic segments. This is a stark contrast to Drosophila, where Dm-Shc only plays a minor role in the receptor tyrosine kinase (RTK) pathways Torso and EGF Receptor (EGFR). However, in Drosophila, two additional adaptor proteins are involved in these two pathways, Dm-Dos („daughter of sevenless") and Dm-Drk („downstream of receptor kinase") that mediate the activation of Torso in a redundant manner together with Dm-Shc. We tested if these three adaptor proteins function redundantly in Tribolium as well. The knockdown of Tc-Dos resulted in embryos with phenotypes much like those of Tc-Shc where the growth zone is completely missing, i.e. also Dos knockdown appears to block Torso signaling entirely, unlike in Drosophila. Both Tc-Shc and Tc-Dos also displayed weak dorso-ventral defects (only visible using DV molecular markers), which suggests that both are involved in the formation of the dorsoventral polarity in oocytes (Gurken pathway in Drosophila). The third factor, Tc-Drk plays an essential role already during early stages of oogenesis much like EGFR. Thus, parental RNAi cannot be used to study Tc-Drk function during embryogenesis. Using embryonic dsRNA injection, we are currently analysing EGFR and Drk embryonic phenotypes. Preliminary data suggest that Drk is the dominant adaptor in EGFR signaling in Tribolium and is essential for maintenance of the dorsoventral polarity of the growth zone, ventro-lateral positioning of the leg primordia, and proximo-distal patterning of the leg. Based on these results it appears that the function of the three adaptor proteins Shc, Dos and Drk in EGFR signaling is similar as in Drosophila while Dos and Shc play more prominent roles in the Tribolium Torso pathway.

P-138 Role of epigenetic changes in generating non-adaptive genomic variability and evolutionary novelty

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Lamarckian and Darwinian theories of evolution are powerful epistemological constructs that differ from each other in one main aspect: Lamarckism focuses on the role of the environment as a direct inducer of evolutionary novelty, while Darwinism (especially neo-Darwinism) assumes that the origin of evolutionary novelty is a random process. The molecular origin of genotypic variability would be interpreted in the realm of Mayr's proximate cause, while the maintenance of these genomic changes would pertain to Mayr's ultimate causes. In Mayr's view, evolutionary biologists would be concerned with the ultimate causes rather than with the proximate causes, which would primarily be the focus of functional biologists. Explanations based on ultimate causes have predominated in evolutionary biology, even though mechanistic evidence for evolution commonly emerges from functional analyses. Studies on proximate

causes of evolutionary change (inquiring into the mechanistic generation of evolutionary novelties) have since long been performed, mainly by Evo-devo studies. However, most Evo-devo research has not incorporated the Lamarckian component of focusing on environmental inputs and their integration into developmental processes leading to phenotype formation. Epigenetic mechanisms fill this gap, by connecting environmental influences with long-term regulation of gene expression. However, and more importantly, epigenetic mechanisms allow the understanding of the origins of genomic change. Recent empirical evidence exists for: (1) the occurrence of non-random, biased-mutations, in particular GC-biased or increased CpG to TpG transitions, (2) the vast majority of genetic changes not being associated with fitness increases, and (3) environmental exposures that induce germ-line genomic and/or epigenomic changes. This contradicts one of the main assumptions of neo-Darwinian theory: that the main source of genetic variability is mutations occurring stochastically and independent from direct environmental influences. Conversely, epigenetically induced genetic variability can fit Kimura's neutral theory of evolution, since biased-mutations will have phenotypic consequences independent of their fitness effects. Thus, environmentally induced epigenetic changes can be an important component in generating genomic variability that could in turn be neutrally maintained in populations (Epinduced Neutral Evo). This highlights the importance for evolutionary biologists of focusing on proximate causes in order to understand the molecular mechanisms responsible for the origin of genetic variability. When the focus of evolutionary questions are the proximate causes rather than the frequently used ultimate causes, then the emergence of non-random, induced genetic variability can be considered evolutionary relevant even when neutral from a fitness perspective.

P-139 Role of mechano-dependent ion channels in pulsational growth of colonial hydroids

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Apical growth in all parts of the colonial hydroids occurs as a result of growth pulsations. Pulsational growth of hydroids is an interesting example of cyclic morphogenetic processes. It is well known that the apical morphogenesis hydroids accompanied by changes in the parameters of growth pulsations and possibly determined by these changes. The investigation of the mechanisms underlying the pulsational growth of hydroids has important implications for understanding the evolution of morphogenesis. We investigated the physiological mechanisms of cell movement pulsational growth, in particular the role of mechanosensitive ion channels that process. As object of research we chose the top of the stolons, as the most simple system which is not subject morphogenesis and convenient methodologically. Colonies of Gonothyraea loveni cultivated on glass slides, and growth pulsations recorded and processed automatically by an original computer program written to track the movements of individual cells. Gadolinium ion is used as a blocker of mechano-dependent ion channels. Gadolinium significantly slowed the growth of the tips of stolons at a concentration of 50 uM and completely inhibited it at 100 uM. The effect is reversible and is abolished after washing gadolinium. The obtained results indicate the involvement of mechano-dependent ion channels in pulsational growth of colonial hydroids.

P-140 Roles of retinoic acid signaling in architecting the nervous system of Amphioxus

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Understanding the evolution, development and function of the nervous system is one of the major challenges in biological research. In vertebrates, the retinoic acid (RA) signaling cascade has been shown to be an important regulator of the complex ontogenetic processes that lead to a functional nervous system. In order to obtain a better understanding of how RA influences the formation of the overall architecture of the nervous system and the development of neurons with different neurochemical phenotypes, we are using the cephalochordate amphioxus (*Branchiostoma lanceolatum*) as a model system. Amphioxus has not only a very important phylogenetic position at the base of the chordate phylum, but is also characterized by a vertebrate-like central nervous system that contains only about 20,000 neurons and by a genome that shares a high level of synteny with vertebrate genomes, without having undergone the typical vertebrate whole genome duplications. Using a combination of pharmacological treatments, gene expression surveys and immunohistochemical analyses, we have characterized the effects of RA signaling on the development of peripheral neurons and several neurotransmitter systems. We find that RA signaling affects the number, the position and the projection pattern of epidermal sensory neurons. Moreover, our experiments show that the differentiation of certain cell groups (e.g., dopaminergic cells) to their specific neurotransmitter phenotype is also regulated by RA signaling. The observed effects differed remarkably depending on the developmental

stage at which pharmacological treatments were applied. Altogether, these data suggest that RA signaling is required at particular stages of neurogenesis to regulate, for example, the overall number and distribution of sensory neurons in the epidermis as well as the differentiation of cell-specific neurochemistry.

P-141 Seeing eye to eye with the spiders: Differential expression of eye development genes in different eyes of *Cupiennius salei*

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It seems that only a limited set of transcription factors has been deployed in the ontogenic development of the eyes across the phyla and that these factors probably have already existed as precursors in metazoans once the Cnidarians diverged. We tested the expression pattern of the candidate genes involved in the eye development in the wandering spider *Cupiennius salei* embryos to profile the molecular development of the eyes and in search of possible clues as to how different eyes differentiate. We screened the spider embryonic transcriptome and found out that several of eye development genes have been duplicated. Our results show that the two orthologs of the genes have different expression patterns. The genes are mainly expressed in the developing optic neuropiles of the eyes (lateral furrow, mushroom body and arcuate body) in earlier stages of development (160-220h after egg laying). Later in the development (180-280h after egg laying), there is differential expression of the genes in disparate eyes; for example Cs-Otxa is expressed only in posterior-lateral eyes, Cs-Otxb, Cs-Six1a and Cs-Six3b in all the secondary eyes, Cs-Pax6a only in primary eyes, Cs-Six1b in posterior-median and posterior-lateral eyes, and Cs-Six3a in lateral and primary eyes. Our data elucidate that the genes involved in the eye development in other metazoans are conserved in spiders however the genes are deployed differentially to differentiate each particular eye type of the spiders. In accordance with the well-established role of Pax6 in bilaterian eye development, our study implicates Cs-Pax6 only in the spider primary eye development.

P-142 ShapeQTL: Mapping multiple loci for multi-dimensional trait in R

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Geometric morphometrics provides very peculiar high dimensional data. The nature of shape data makes compulsory the use of multivariate approaches: (1) reducing ourselves to the analysis of only one composite trait is not fully satisfactory because nothing ensures that the genetic variation is the main player and that accordingly structures adequately the shape space; (2) the stack of results from univariate mapping of each coordinates misses proper multivariate testing whereas this test will be generally more powerful than the univariate ones. This increase in power is not the sole interest of using multivariate mapping, high dimensional traits have also some interesting feature against long range smoothing due to the high linkage found in inbred crosses because the q-dimensional effect will quickly move away as probabilities of the QTL genotypes change, property that should add power to identify linked QTLs. I present an R implementation of Haley-Knott regression that handle reduced-rank data and can be apply to shape data as well as other multivariate traits. The program is built on the R/qtl implementation and extends its stepwise model search based penalized LOD scores to multidimensional traits.

P-143 Shavenbaby functions as a segmentation gene in the short germ embryo of Tribolium and may be regulated by mille-pattes

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In short germ insects like *Tribolium castaneum*, the abdominal gap genes appear to have undergone significant evolutionary divergence in function and regulatory interactions. Of particular interest is the novel Tribolium gap gene mille-pattes (mlpt) whose ortholog in Drosophila (polished rice/tarsal-less; pri/tal) has no function in segmentation despite a strong similarity in deduced amino-acid sequence and gene structure. The mlpt phenotype is characterized by a loss of abdominal segments, their transformation to thoracic identity, and the loss of telson structures. As in pri, the mlpt locus encodes a polycistronic mRNA that codes for several very short peptides. In Drosophila the Pri peptides regulate trichome pattern by inducing proteolytic cleavage of the zinc-finger transcription factor Shavenbaby (Svb), converting it from a transcriptional repressor to a transcriptional activator. We find that the *Tribolium* ortholog of svb (Tc-svb) plays a role in abdominal segmentation, as knockdown of this gene results in a phenotype similar to mlpt, with missing abdominal segments, and their homeotic transformation to thoracic identity. However, differences in knockdown phenotypes, expression, and regulatory interactions suggest that repressor and activator variants of Svb function in segmentation. While the expression domains of mlpt and Tc-svb overlap only partially, the boundaries of their expression domains are in close proximity indicating an interaction between Mlpt and Tc-Svb that may involve a non-cell autonomous effect of Mlpt. Using a heat-shock overexpression system, we have shown that the Mlpt peptides regulate segmentation and segmental identity at various stages during

embryonic development. One late effect of mlpt overexpression is a strong telson transformation phenotype that involves misregulation of the HOX gene, Abdominal-B. This specific overexpression phenotype can be employed as a valuable assay for analyzing the function of the Mlpt peptides.

P-144 Snakes and amphisbaenians share molecular signatures in the Conserved Element B, a regulatory fragment for terminal HoxD expression during vertebrate development

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The evolution of snakelike phenotypes, which are characterized by elongated trunks and absent limbs, has occurred independently many times among Squamata. HOX genes are crucial for coordinating the development of different structures in vertebrate embryos, and variations in their expression patterns often relate with morphological diversification. Variations in gene expression patterns may be triggered by nucleotide mutations in gene regulatory regions, which affect cis-trans interactions. The genes HoxD10 to HoxD13 are expressed during development of autopodium, axial skeleton and genitalia in the tetrapod embryo, and the conserved element B (CsB) within the Global Control Region plays a regulatory role as 5′ HoxD enhancer. We investigated if the independent evolution of a snakelike morphology in two squamate lineages (Serpentes and Amphisbaenias) involved equivalent molecular signatures in CsB. Conserved fragments of CsB (B1 [725 pb] and B2 [423 pb], 1148 pb total) were cloned and sequenced from 15 snakes, 14 amphisbaens, eleven lizards and one crocodile; sequences from chicken, one turtle and two mammal species were retrieved from GenBank for additional comparisons. Transcription factor binding sites (TFBS) were predicted using match algorithm and the TRANSFAC database. Comparisons between Squamata (lizards, snakes and amphisbaens) and the remaining species reveal variation in the predicted TFBS distributed throughout almost the entire segment (from position 133 to 1074). Among Squamata, however, variation seems restricted to B2 (735-1062). Snakes present a higher amount of exclusive predicted TFBS in comparison with other squamates (756-1062), whilst differences are concentrated in a smaller portion of the fragment in amphisbaens (790-850). Besides variations in CsB sequences detected as being exclusive to each lineage, the two groups characterized by a snakelike morphology share some TFBS signatures when compared to lizards, which are located in a short segment (841-849): they apparently lost binding sites in specific positions for c-Maf (841), HIC1 (843), NeuroD or TTF-1 (847), AP-4 (848) and XPF-1 or LBP-1 (849). Additionally, a HIC1 TFBS in lizards seems to have been replaced by a PBX1 TFBS in the snakelike phenotypes at position 846. PBX1 is relevant for axial skeletal development, and its gain in snakelike groups CsB might have played a role in evolution of elongated trunks. The TFBS in positions 841-849 may be functionally important during limb development, and their functional loss in limbless species may have culminated in the substitution for other TFBS.

P-145 Springtails as basal hexapod models for comparative genetic studies

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Springtails (Collembola) are one of the basal-most lineages of Hexapoda, which is a group of arthropods that also includes the insects. A number of arthropod species are being used for Evo-Devo genetic research, but genetically tractable models are still missing within the basal hexapods. Such models would be beneficial for example, for the understanding of the yet enigmatic origins of the hexapods. To fill in the gap we are introducing the springtail *Orchesella cincta* for comparative genetic studies. *Orchesella* has long been used in ecotoxicology by researchers at the University of Amsterdam, who established the culture. We will present the advantages and current limits of *Orchesella.* The main advantages include functional systemic RNAi. We will demonstrate this using the example of how Hox genes control the development of a characteristic springtail trait — the abdominal appendages.

P-146 Steroid-signalling evolution: The Lophotrochozoan ecdysone receptor

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Nuclear receptor (NR)-mediated signalling regulates various biological processes such as development, physiology and reproduction. They belong to a diverse superfamily of ligand-dependent, or independent, transcription factors that participate in the homeostatic regulation of hormonal systems through gene expression modulation. This diversity is, indeed, reflected by their ligand-binding affinities: expanded and fine-tuned during metazoan evolution. Apart from the classical physiological roles, NRs clustering in the sterol-binding branch have been suggested to participate in the modulation of longevity in different invertebrate species: the C. elegans Daf-12 receptor, shown to ligate a bile acid-like steroid, and the *D. melanogaster* Ecdysone Receptor (EcR), activated by ecdysteroids, also known as moulting and reproductive hormones. Of peculiar interest is the suggested

phenomenon opposing development and normal reproductive growth versus latency and extended longevity. Ecdysteroid signalling acts via a heterodimeric nuclear receptor complex formed by the Ecdysone Receptor (EcR) and the Retinoid X Receptor (RXR), or its insect homolog Ultraspiracle (USP). While RXR orthtologues have been found in virtually all the bilaterian species studied, EcR homologs have been only isolated from arthropods and from the phylogenetically-related nematodes (Ecdysozoans). Yet, we have identified and cloned a Lophotrochozoan EcR orthologue: corroborated by genome database mining. Along with this finding several questions arose. Are Lophotrochozoan EcR receptors functional and ligand-activated? Given that non-arthropod invertebrates seem unable to de novo synthesize these steroid hormones, what are their ligands? Finally, if functional, different scenarios can be proposed taking in account the physiological role of ecdysteroids in Ecdysozoans: development, reproduction and/or longevity. To understand the physiological significance of a Lophotrochozoan EcR we address the evolutionary history and molecular function of this receptor. The molecular characterization of the lophotrochozoan EcR will be followed by gene expression pattern analysis in adult tissues and during development.

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P-147 Structure, function, conservation, and evolution of C2H2 zinc finger transcription factors in arthropods

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With gene regulation as one of the major themes in evolutionary developmental biology, research is focusing both on non-coding *cis*-regulatory sequences, and on the transcription factors that bind them. While some families, like homeobox-containing transcription factors, have been extensively studied, others have received less attention. The C2H2 zinc finger gene family is one of the largest families of transcription factor genes, with important roles in many developmental processes. Yet, while many zinc finger genes have been researched individually, the family as a whole has not been studied in an evolutionary context. We have attempted to address this gap by analysing all zinc finger-containing transcription factors in a sample of currently sequenced and annotated arthropod genomes, looking at the number and type of zinc fingers, as well as other conserved motifs. Our analysis confirms the existence of distinct groups of zinc finger proteins, identified by a typical combination of eight distinct zinc finger domains. Furthermore, using available protein databases, we ask whether individual zinc finger domains can be assigned to particular functions, both on the protein level and with regard to specific developmental processes. Looking at the different groups of zinc finger proteins, we investigate how conservation of function is linked to the conservation of gene architecture. We hypothesize that genes with a highly conserved structure have been under stabilising selection to retain ancestral roles, whereas gene groups portraying a more variable structure also have variable functions and are probably under lineage-specific selection.

P-148 Super-size me: On the quest for increasing molar size while maintaining shape

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The scaling of patterning — the mechanisms through which organs of different-sized animals attain the correct forms and functions — has lacked rigorous analysis. Especially interesting is the question of how organ size increases while maintaining the same shape. In principle this problem can be solved in two ways; by additional growth after patterning events, or by attaining shape at a larger size, which in many cases would require changes in the patterning mechanisms. Mammalian molar teeth develop as a result of a series of morphogenetic movements and signaling interactions between ectodermal epithelium and neural crest-derived mesenchyme. The molar cusp patterns are preceded by the appearance of secondary enamel knots at the tips of the future cusps. These signaling centers regulate cusp formation. We compared molar development in two rodent species with similar tooth shape, but a twofold difference in length: house mouse (*Mus musculus*) and brown rat (*Rattus norvegicus*). We used microcomputed tomography and in situ hybridization to follow the development of molar size and shape. Our data shows that rat molar attains its shape at a larger size compared to that of the mouse. In addition, preliminary results suggest that increasing tooth size while maintaining the shape requires an increase in two factors: the tooth bud length and the spacing between the secondary enamel knots.

P-149 *Sycon ciliatum* (Calcarea, Calcaronea) regeneration peculiarities

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The ability to regenerate is widespread in the animal kingdom, but the regenerative capacities and mechanisms vary widely. To understand evolutionary history of the diverse regeneration mechanisms, regeneration processes must be studied in early-evolved metazoans along the traditional cnidarian and bilaterian models. For this purpose, we have combined several microscopy techniques to study mechanisms of regeneration of the calcareous sponge, *Sycon ciliatum*. The objectives of this work are to detect the cells involved in this process and to reveal the morphogenetic processes, with a special interest in recovery of axial polarity. *S. ciliatum* exhibits a radial symmetry with elongated choanocyte chambers radially arranged around a central atrial cavity and clear apical-basal (AP) polarity. When the sponges are cut perpendicular to their AP axis, resulting in a series of "body rings", each of the rings has a capacity to regenerate the amputated basal and apical parts, while preserving their original axial polarity. The cutting exposes choanocytes, the inner epithelial cells responsible for generating of the water movement and capturing of food particles. Between 6 and 12h after the cut, all cells in the exposed area ("wound zone") and around this zone show active movement, dedifferentiation and transdifferentiation. At the same time, pinacocytes migrate to the wound as a continuous sheet. At 24h the main wound surface is covered with pinacoderm, complete with new ostia (incurrent openings). Subsequently, the atrial cavity becomes covered by membrane, composed of new exopinacoderm and endopinacoderm, which assemble in internal border zone. This membrane closes completely at the basal end of the regenerating ring, but remains open and forms oscular sphincter at the apical pole. There are three main sources of the new pinacoderm produced during regeneration: intact exopinacoderm, intact endopinacoderm, and significantly, intact choanoderm of the wound surface. The basic morphogenetic processes during regeneration are spreading (flattening) and fusion of epithelial sheets. The other intriguing mechanism is the transdifferentiation of choanocytes into exo- and endopinacocytes. We have not observed epithelial-mesenchymal transitions during regeneration. No increase in the cell proliferation was detected during regeneration, and thus the regeneration in *S. ciliatum* is morphallactic: lost body parts are replaced by the remodeling of the remaining tissue, accompanied by transdifferentiation of the cells. The transdifferentiation capacity of choanocytes and their involvement in regeneration supports the notion that these cells combine features of somatic and stem cells.

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P-150 Temporal shift and axis specification during the evolution of early vertebrate development

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Gastrulation in birds and mammals is accompanied by formation of the primitive streak as an evolutionary novel site of epithelio-mesenchymal transition and results in germ layer development including the emergence of the notochord from the area of the primitive node. Primitive streak formation in amniotes, although starting from different topographical conditions, has been suggested to emerge twice and independently from the ancestral blastopore and is, therefore, a candidate for a developmental constraint. The flat embryonic disc of the rabbit exhibits extensive lateral to medial tissue rearrangement and elongation prior to gastrulation suggesting a heterochrony of cellular movements similar to the one discussed for the chick embryo. We inhibited this temporal shift experimentally in the rabbit and thereby produced novel gastrulation forms resembling possible steps of the transformation of a circular blastopore into a straight primitive streak. Another example of a temporal shift causing a divergence of fundamental developmental mechanisms may be left-right symmetry breaking: Expression of nodal mRNA in the left lateral plate mesoderm soon after the beginning of somitogenesis is a common feature in all studied vertebrates but developmental processes prior to this asymmetry vary; in most studied model organisms symmetry breaking is associated with ciliary flow at the so-called left-right organizer but this seems not to be the case for chick and pig embryos. Moreover, in the chick the asymmetry of the paraxial nodal domain can be considered to occur "precociously" as it is temporally shifted to the beginning of notochord formation, whereas paraxial nodal domains in mouse, rabbit and Xenopus start in a symmetrical fashion. We propose a novel sequence of evolutionary modifications of gastrulation topography and discuss the role of heterochrony and developmental constraints in early vertebrate development.

P-151 Terminal differentiation in reaction-diffusion models

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Reaction-diffusion is commonly applied in the modelling of biological pattern formation. While these models can potentially greatly improve

our understanding of the patterning process under study, or help testing specific hypothesis, the biological relevance of the models is often questioned due to underlying simplifying assumptions. One such major simplification in classical reaction-diffusion models is that the model domain remains static over time, and the patterning is established as a result of a balance of morphogen concentrations. We investigate, by simulation, the role of cell terminal differentiation as a mechanism to augment the pattern generation capabilities of classical reaction-diffusion models. In such a model the morphogen concentrations in cells cause irreversible state changes in cell properties, such as morphogen level limits or secretion rates. Terminal differentiation as a process is highly essential in pattern formation in nature, thus including terminal differentiation in a reaction-diffusion model is well founded. We also briefly discuss how the choice of a particular numerical scheme and the spatial resolution of the model domain could affect the model outcomes in non-desirable ways. Reaction-diffusion models are typically implemented without much consideration to these questions, which could potentially lead to problems with the interpretation of the results.

P-152 Testing the role of amniotic marker genes on (extra)embryonic development in the red flour beetle, Tribolium castaneum

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Formation of the extraembryonic membranes (amnion and serosa) in insects is an important process for the proper development of the entire embryo. Therefore, the investigation of genes that are expressed in specific domains known to give rise to those membranes should provide new information about their specification and lead to a better understanding of the associated developmental processes. During early extraembryonic development (e-EED) in the red flour beetle, *Tribolium castaneum*, the embryo and amnion together invaginate into the yolk, while simultaneously the serosa starts surrounding both of them. When the serosa completely lines the inner eggshell, the embryo undergoes further germband extension, with its ventral surface covered by the amnion. At this point, formation of the two membranes is complete, and the embryo is now fully enveloped and protected by the amnion and the serosa, where the latter epithelial membrane additionally surrounds the yolk. Prior to the morphogenetic movements of e-EED, during the differentiated blastoderm stage, a one to two cell wide oblique row of cells marks the border between serosa and germ rudiment. These cells express both iroquois (iro) and pannier (pnr), two commonly used amniotic marker genes. By silencing both genes via RNA interference, I examined the resulting phenotypes in regard to a possible role in amniotic tissue specification. Although data for iro are still under evaluation, our surprising conclusion is that neither early amniotic marker seems to fulfill such a role. Instead, loss of Tc-pnr expression leads to defective dorsal cuticle formation and a partial loss of cardioblast cells. These results are in line with the findings in the fruit fly *Drosophila melanogaster*, where this dorsal opening is seen in pnr deficient mutants. Furthermore, pnr, together with tinman, induces the cardial cells in *D. melanogaster*. Further extension of these experiments to the milkweed bug *Oncopeltus fasciatus* (Hemiptera), will provide an evolutionary perspective on how conserved the genes are and insights into the other main mode of extraembryonic development across the insects. The „emerging model organism" status of *T. castaneum* includes the availability of several transgenic lines that contributed to the analyses described above. To augment and refine our capacity for tissue specific visualization and mis-expression, I am working on a system that will facilitate the easy insertion of a new transgene into targeted genomic sites by addition of a phiC31 integrase binding site, facilitating future manipulations under the control of specific enhancers.

P-153 The development of palate of the miniature pig

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Clefts of the lip and/or palate (CL/P) are among the most common birth defects worldwide. Orofacial clefts occur with a frequency of 1 to 2 per 1000 live births and cleft palate accounts for 30% of orofacial clefts. The majority of CL/P are non-syndromic where CL/P occurs in isolation of other phenotypes which is caused by the failure of the secondary palatal processes — medially directed, oral projections of the paired embryonic maxillary processes — to fuse. Both gene mutations and environmental effects contribute to the complex etiology of this disorder. To better understand the development of palate, we investigated the development of the palate of miniature pig as big animal models. We found that the palate was formed from E25 to E50. At E25, lateral palatine processes began downward on either side of the tongue, separating the tongue and oral epithelium. By E30, the palate shelves began to ascend and turn to horizontal direction, due in part to the downward movement of the developing tongue, removing a physical barrier to palate elevation. At E45, the palatal shelves began to fuse one another. And at E50, the palatal shelves approached one another, resulting in a complete separation between the oral and nasal cavities. Compared with the mouse, whose palate development formed at the middle-end stage of the embryonic period,

the development of the palate of the miniature pig was in the early-middle stage during embryonic period. The development stage and structure of pig palate was closer to human.

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P-154 The echinoderm larval skeleton as a possible model system for experimental evolutionary biology

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The evolution of various body plans results from the acquisition of novel structures. Some novel structures necessitate multiple evolutionary steps, requiring organisms that overcome the intermediate steps that might be less adaptive or neutral. To examine this issue, echinoderms might provide an ideal experimental system. A larval skeleton is acquired in some echinoderm lineages, such as sea urchins, probably via the co-option of the skeletogenic machinery that was already established to produce the adult skeleton. The acquisition of a larval skeleton was found to require multiple steps and so provides a model experimental system for reproducing intermediate evolutionary stages. We also present the result of experimental reproduction of the intermediate stage, and discuss how the echinoderm overcomes the intermediate step. We also present evidence that is not consistent with the long accepted idea of convergent evolution of pluteus larva in sea urchin and brittle stars.

P-155 The effect of floral variation in the field bean (*Vicia faba*) on pollinator behaviour

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Phenotypic variation and the genes underlying it are the basis upon which selection acts. This is not only true in natural systems, but also in artificial ones, such as seen in agriculture. The field bean (*Vicia faba*) is an important legume crop with a high protein content and ability to fix nitrogen. However, it requires pollination by bees to achieve maximum yield. In recent years, the yield of *V. faba* on a field-to-field basis has become more variable. With declining bee populations it is becoming increasingly important to optimise pollination of this crop. A promising and little studied avenue for improvement of the yield and yield stability of *V. faba* is to target floral traits. By determining how the natural variation in floral traits affects bee behaviour in this crop, it should be possible to improve pollination rates (assuming the optimum has not already been unconsciously selected for). This will lead to enhanced yield and yield stability. Few studies have attempted to investigate the effect of floral variation on pollinator preference in *V. faba*, and none comprehensively. Therefore, the variation of floral traits in inbred lines of *V. faba* has been assessed under standardized environmental conditions. Five main groups of traits that have been shown to affect pollinator preference are being investigated: floral colour, petal epidermal morphology, general floral shape, reward provided and volatile profile. Variation has been identified in each of the traits that have been assessed so far. Future work will now take a two-pronged approach, both identifying the genetic basis of key floral traits, and examining how this variation affects pollinator response using preference tests with *Bombus terrestris* under laboratory conditions.

P-156 The effect of oxygen deficiency on early ontogenesis of common toad (*Bufo bufo*)

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The influence of oxygen deficiency on the survival and developmental rate of common toad embryos was experimentally studied. In total, 7200 fertilized eggs from four clutches were used. The experiment was carried out under conditions of low and high egg density (30 or 120 eggs per 0.1 l). Eggs were split up into four groups developed under the following conditions: oxygen deficiency and low density; oxygen deficiency and high density; normal oxygen supply and low density; normal oxygen supply and high density. The experiment has been finished at the hatching stage. Concentration of dissolved oxygen was measured in the beginning and at the end of the experiment with a digital optical sensor LDO of the oxymeter HQ30D.99 (HACH). At the end of experiment, the oxygen concentration was significantly lower in aquariums with the oxygen cut off than in the aquariums with easy access to air. It was shown that the total mortality rate was significantly higher under the oxygen deficiency than under the normal oxygen supply only in the group developed under high density. The development of embryos slowed and stopped at the gastrula stage; then the mass death of embryos began, and only 2-3% of embryos survived until the hatching stage. In the groups developed under low density, the total mortality rate was significantly high under the oxygen deficiency only in 2 of 4 clutches investigated. The survival rate in the low density groups varied from 64 to 96% and the developmental rate of embryos slowed down under the oxygen deficiency. However, the developmental rate in these groups was significantly higher than in the high density groups. Earlier, in experiments with the culturing

of single embryos, I showed that oxygen deficiency does not lead to increase of total mortality rate, but shiftsthemortality to earlier stages of development (Dmitrieva 2008). Thus, oxygen concentration imposes different effects on mortality and developmental rate of toad embryos developing under the different densities. Theoxygen deficiency has a greater effect on the embryos developing under high density.

P-157 The evolution of floral traits in the Antirrhineae

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Flower evolution has been an important force behind angiosperm diversification. In recent years the function of several genes involved in flower development has been elucidated; however little attempt has been made to match this information with the ecological and evolutionary profiles of plant groups. Due to its morphological and ecological diversity, the tribe Antirrhineae, which includes model species *Antirrhinum majus,* is a good system to study floral trait variation. In this project, we are looking into the evolution of three traits: petal epidermal cell shape, nectar storage receptacles and corolla shape using a combination of morphometric and molecular biology techniques. We have analysed the expression of MIXTA and its homologues: MIXTA-like1, MIXTA-like2 and MIXTA-like3, responsible for the development of conical cells. Likewise the role of KNOX genes such as Invaginata and Hirzina as potential regulators of nectar spur formation is being investigated. Finally, corolla shape has been characterised using an allometric model based on landmarks and analysed using Principal Component Analysis (PCA).

P-158 The evolution of the RDH10 gene family: Duplication and lineage-specific loss of a novel member

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Nuclear receptor-mediated retinoic acid signalling is crucial for organ modelling and maintenance. Stored and free retinol serve as precursors for the biosynthesis of active retinoids: a two-step oxidation cascade leading to signal transduction. Retinol is first oxidized into retinaldehyde, in a reversible manner; renitaldehyde is, in turn, irreversibly oxidized into retinoic acid. While the first metabolic reaction was initially thought to be ubiquitous and unregulated, the spatiotemporal coordination of retinoic acid supply was attributed to the retinaldehyde-to-retinoic acid conversion. Yet, recent studies have highlighted the functional role of a membrane-associated retinol dehydrogenase (RDH10) in the first oxidation step in mammals. This enzyme displays tissue and time-specific expression patterns that correlate with both retinoic acid and retinaldehyde dehydrogenase activities, suggestive of an additional checkpoint for retinoic acid regulation. In this study we investigated the evolution of chordate rdh10. While a single copy, rdh10a, is observed in birds and mammals; reptiles, amphibians, teleosts and chondrichthyans exhibit an additional uncharacterized gene, rdh10b. Both rdh10a and rdh10b have additional duplicate copies in teleosts. Phylogenetic and paralogy analysis revealed that vertebrate rdh10a and rdh10b resulted from whole genome duplication in stem vertebrate evolution. Following duplication, rdh10b was lost in warm-blooded lineages and retained in most cold-blooded animals. Both enzymes exhibit conserved reaction cores and tri-dimensional folding; yet, the membrane-association designs appear different: unlike RDH10A, topology predictions advocate for RDH10B solubility. Also, a unique negatively charged insertion is observed in RDH10A isoforms. Finally, we also addressed the gene expression patterns of rdh10a and rdh10b in teleosts, *D. rerio* and *O. nicotilus*, both during development and in adult tissues. Taken together these results support a functional specialization within the rdh10 family and suggest a dichotomy among vertebrates according to thermal homeostasis mechanisms.

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P-159 The evolutionary origin of the vertebrate midbrain

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The evolutionary origin of the vertebrate midbrain remains enigmatic, though it is one of the most important neural region as a integrative brain centre for non-mammalian vertebrates. Image-forming vision is one of the main functions of the midbrain, but our research comparing nervous system development between amphioxus and lampreys revealed that the ancient visual centre of chordates was in the prosencephalic region. In addition, the image-forming vision was acquired in the vertebrate lineage as a result of rearrangement of the visual centre from the prosencephalon to the midbrain. These results indicate that the midbrain was acquired in the common ancestor of vertebrates. This speculation superficially disagrees with the existence of the midbrain-hindbrain boundary (MHB), or isthmic organiser, in protochordates. For explaining the evolution of midbrain,

it is necessary to consider not only the genetic regulatory network (GRN) but also the neural or developmental function of the ancestral midbrain and isthmic organiser. Here we discuss the evolution of the midbrain based on results of our recent experiments.

P-160 The evolutionary origins of vertebrate blood cells

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A challenge for evolutionary and developmental biology is to reveal the genetic mechanisms underlying the origin of evolutionary novelties. Most bilaterians have a basic hematopoietic system that mainly produces innate-immune cells; however, vertebrates also have highly specialised cells such as erythrocytes and thrombocytes. This astounding diversity of blood cell lineages was a key evolutionary innovation for vertebrate evolution. Their appearance coincides with two rounds of whole genome duplications at the base of the vertebrate lineage. These events may have contributed to the establishment of multiple blood cell lineages. Amphioxus, the basal chordate, did not undergo the whole genome duplications, and thus can represent the vertebrate ancestor. In adult amphioxus, immune cells have already been identified after challenge with bacteria. Moreover, recent study has shown that amphioxus embryos have haematopoietic cite homologous to vertebrate AGM region. To further analyse haematopoietic development in amphioxus, and better understand the molecular changes behind the evolution of the vertebrate multiple blood cell types, we have identified and cloned amphioxus genes homologous to those involved in the regulatory network of vertebrate haematopoiesis. So far, we have analysed the expression of some of these genes in amphioxus embryos and the functional conservation of amphioxus Gata1/2/3 gene in zebrafish embryos.

P-161 The expression of *Fzd6* in the dental lamina of monophyodont and diphyodont dentition

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Frizzled 6 (Fzd6) belongs to a family of proteins that serves as a receptor in Wnt signalling pathways. It acts as a positive regulator of the noncanonical Wnt and a negative regulator of the canonical Wnt/β-catenin signalling cascade. Fzd6 plays an important role in the establishment of planar cell polarity in fly wings or hair follicle in mice. The aim of our study was to determine if Fzd6 controls dental lamina asymmetrical growth into the mesenchyme. We compared several embryonic developmental stages in two animal species — the mouse with monophyodont dentition and the pig with diphyodont dentition. In the pig, a gentle positivity was visible in the epithelial thickening of dental lamina (E25). Next, we found noticeable asymmetrical expression of Fzd6 in the oral epithelium with distinctively stronger signal on the labial side of the dental lamina and weaker expression in the budding tissue. At later stages (E36, E56, E67), the expression was more distinct in the main body of the dental lamina including cells separating from the lamina during its regression. In the mouse, the expression pattern was similar during early odontogenesis. Fzd6 expression was located in the oral epithelium on the labial side of the dental lamina and weak expression was observed in the dental tissue at early stages (ED15.5). Later in development, the expression was situated in the lingual side of the dental lamina (ED17.5). In conclusion, we found asymmetrical expression of Fzd6 in the dental lamina as well as in the oral epithelium of early stages. This asymmetrical expression may be important in growth directionality and side-specific morphological differences of the dental lamina.

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P-162 The extraembryonic serosa protects insect eggs against microbial infection and other ecological impacts

Jacobs, Chris (Leiden University, NLD); **van der Zee, Maurijn** (Leiden University, NLD)

All major arthropod groups have colonized land to some extent, but insects have been the most successful in the invasion of terrestrial habitats. This ecological and environmental transition from an aquatic lifestyle coincided with the origin of an evolutionary novelty in insect eggs: the serosa. The serosa is an extraembryonic epithelium that envelops the embryo and yolk. We have previously shown that the serosa provides desiccation resistance to the egg and might have facilitated the spectacular terrestrial radiation of the insects (Jacobs et al. 2013). We now demonstrate that the serosa is also required to mount a potent immune response. In contrast to Drosophila eggs, eggs of the flour beetle *Tribolium castaneum* exhibit a full-range innate immune response involving Toll and IMD signaling, the production of antimicrobial peptides (AMPs), melanisation, and the production of reactive oxygen species. When we delete the serosa by applying parental RNAi against Tc-zen1, this response is almost eliminated. We further show expression of crucial bacterial recognition genes in the serosa. Thus, we propose that the serosa is a frontier epithelium that protects insect embryonic development against ecological and environmental impacts, such as desiccation and microbial infection. A

trade-off with developmental speed might have driven the loss of the serosa in a small group of derived Diptera to which Drosophila belongs.

P-163 The function of Oct4 homologues in the evolution of epiblast versus germ cell potency: Relevance to embryonic stem cell self-renewal and induced pluripotency

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Oct4 or Pou5f1 is a master regulator of pluripotency and differentiation in both embryonic stem cells (ESCs) and during embryonic development. It has conserved roles in gastrulation stage differentiation (epiblast activity) and germ cell specification (germ cell activity). Based on the expression patterns of different Oct4 homologues (POU5F1, 3) in African clawed frog (*Xenopus laevis*, Xl) and tammar wallaby (*Macropus eugenii*, Me), Oct4 homologues appear to have adapted lineage specific roles. Xlpou91 (pou5f3.1) and MePOU5F1 are expressed specifically in germ cells (germ cell specific POUV proteins), while Xlpou25 (pou5f3.2) and MePOU5F3 are expressed at high levels in gastrulation stage epiblast or ectoderm (epiblast specific POUV proteins). Here we ask if these expression patterns correlate with distinct functional activities in mammalian cell culture. We found that germ cell specific POUV proteins have the capacity to support self-renewal in Oct4-null murine ESCs, whereas epiblast-specific POUV proteins have only a limited ability to support self-renewal, and allow for differentiation to both trophoblast and primitive endoderm. While all Oct4 homologues appear to function in the reprogramming of fibroblasts to induced pluripotent stem cells (iPSCs), we observed marked differences in efficiency. We found that reprogramming efficiency could be augmented by either increasing POUV dose or by giving cells more time to reprogramme. The specific activity of POUV proteins in reprogramming appeared to correlate with their expression in the germ cell lineage. Taken together, our observations suggest that Oct4 has two distinct functions in development that have repeatedly become segregated in evolution: an epiblast function in supporting gastrulation stage progenitors and an independent function in germ cell specification. It is the germ cell specific activity of POUV proteins that best resembled the state present in ESCs and required to promote reprogramming to iPSCs.

P-164 The genome of the cephalochordate *B. lanceolatum*: A step into chordate functional genomics

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The cephalochordate Branchiostoma lanceolatum is a pivotal model for studying the evolution of development and genomes in animals. The availability of the genome sequence for this species is therefore not only a great opportunity to expand the experimental capabilities of the model, but also to extend comparative genomics approaches across chordates and vertebrates. We used Illumina technology to generate a deep sequence coverage (150x) using short insert and jumping libraries. To perform an accurate assembly, we had to tackle the high genome polymorphism in this species using a combination of stringent assembly parameters and haplotype reconciliation. We obtained a high quality draft assembly (N50: 1.39Mb) that was used as a starting point for comparative and functional genomics. In particular, we are developping approaches for extensive RNAseq and Chip-seq based characterization of the regulatory landscape during the development of this species, which will provide an unprecedented opportunity to identify conserved regulatory mechanisms across metazoan phyla.

P-165 The nervous system of Xenacoelomorpha: A tale of progressive cephalization

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According to what has been suggested in the latest phylogenetic studies, the clades Nemertodermatida, Xenoturbella and Acoelomorpha belong to a single monophyletic group named Xenacoelomorpha. Even though most authors consider this group to be the sister of the remaining Bilateria, it has been also suggested that they are a phylum within the deuterostomes.
Due to the relative phylogenetic positions of its constituent three clades, Xenacoelomorpha becomes an interesting animal group for evolutionary analysis (trends). The phylogenetic relationships within the group suggest that Xenoturbellida is the earliest branching group followed by the Nemertodermatida and the Acoela. Tracing the genomic variations associated to the divergence of these groups can give us an idea of how the genomes of these groups have changed over evolutionary time and how these changes relate to their different morphologies. In the last years, several research groups (including

ours) have sequenced the genomes of different Xenacoelomorpha species, studies that allow us to study the evolutionary dynamics of different families of genes. In addition, during this period, several molecular tools have been developed for the better mapping of structures and tissues in these organisms. The organization of the nervous system has often been considered an important phylogenetic character to study. Analysing the development of the nervous system in the Xenacoelomorpha could give us new insights into the early organization of the bilaterian nervous system. With the main aim of studying the origin of the "cephalized" nervous system, we have started to analyse the evolutionary history of different superfamilies of genes linked to the formation and the activities of the nervous system (here the families described are: basic-HLHs, GPCRs and Wnts). We characterize the complements of genes belonging to these families in the acoel *Symsagittifera roscoffensis* and also in *Xenoturbella blocki*.

P-166 The neuroendocrine roles of ventral veins lacking: Is the transcriptional regulation of sexual maturation conserved in metazoans?

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Although endocrine changes are known to modulate the timing of major developmental transitions in metazoans, the transcriptional regulators underlying these changes remain poorly understood. POU transcription factors have been shown to regulate the neuroendocrine changes associated with puberty. In this study, the function of a homolog of the vertebrate POU domain protein, Ventral veins lacking (Vvl)/Drifter, was examined in the red flour beetle, *Tribolium castaneum*. RNA interference-mediated silencing of vvl expression led to both precocious metamorphosis and inhibition of molting in the larva. In insects, two developmental hormones, juvenile hormone (JH) and ecdysteroids, play crucial roles in mediating developmental changes associated with metamorphosis and molting. We show that vvl is expressed in the presumptive endocrine gland precursor cells in embryos and that it plays a critical role in regulating the biosynthesis of both of these hormones during postembryonic development. Our findings show that POU factors modulate the production of major neuroendocrine regulators in both vertebrates and insects. They further support an intriguing hypothesis that the transcriptional control underlying sexual maturation may have an ancient origin in the common ancestor of protostomes and deuterostomes even though the hormones they regulate likely evolved independently.

P-167 The origin of the avian carpal elements, clarifying anatomical confusion

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The homologies of the carpals of the avian wing remain unresolved in comparative development and anatomy. Though the homology and development of the chicken wing has been studied at least since 1864 when Carl Gegenbaur studied the development of vertebrate forelimbs (reviewed in Richardson 2012). Traditionally these questions have been answered using generally 2D methods, pictures of whole-mounts or serial sections, unwieldy for the small and 3D nature of the developing elements. Using 3D confocal microscopy to analyze the mesenchymal condensations and their relationship with the vasculature, we obtain a more accurate picture of the cellular precursors of these elements and better assign their homology and unify their relationship to other tetrapods. Lastly, The origins of the carpal elements help the understanding of the confusing intermediate tissues from which these elements emerge.

P-168 The presence of Vent genes in neural tissues of chordates

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Vent transcription factors are well-known ventralizing homeobox genes that play a crucial role during early specification of dorsoventral axis in chordates. Although vent genes are strongly expressed in neural tissues of chordates, their function in the establishment of neural cell fate is not studied. The expression of Vent genes at the edges of neural plate is conserved among chordates. We show that during neurulation amphioxus Vent1 and Vent2 genes are also expressed in the individual cells of ventral ectoderm, which may represent previously described migrating cells. Additionally, we observed the activity of amphioxus Vent2 promoter in the neural crest of chicken embryos. Our data suggest that the function of Vent genes in the neural development might be ancestral within chordates.

P-169 The reptilian transcriptomes v2.0: An extensive resource for Sauropsida genomics and transcriptomics

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Despite its remarkable diversity, the Class Reptilia remains largely under-represented in major sequence databases and comparative genomic/transcriptomic studies. Here, we use a single pipeline (LANE runner v2) to annotate new transcriptomic and genomic data, as well as multiple published datasets, and build an integrated single resource:

The Reptilian Transcriptomes Database v2.0 (http://www.reptilian-transcriptomes.org). This resource includes representatives of each of the four extant reptilian orders: (1) six Squamata, including three snake and three lizard species; (2) the Sphenodon; (3) three Crocodylia species; and (4) one turtle (Testudines). LANE runner v2 integrates an improved annotation pipeline based on iterative BLAST+ searches and Reciprocal Best Hit (RBH) identification. This approach allows us to annotate a higher percentage of sequences than in previous studies. We also built the so-far largest protein alignments (above 500,000 amino acids per species) for reptiles resolving the position of turtles and the tuatara. The Reptilian Transcriptomes Database v2.0 is a new resource that can serve as a reference for expression analyses, as well as linkage mapping, comparative genomics and phylogenomics. The LANE runner v2 software pipeline can easily be used for the annotation of any transcriptomic dataset. Both LANE runner v2 and The Reptilian Transcriptomes Database v2.0 are publically available in July 2014 at http://www.reptilian-transcriptomes.org.

P-170 The role of the BMP and Toll/NF-kB Pathways in patterning the dorsal-ventral axis of the jewel wasp, *Nasonia vitripennis*

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Bone Morphogenetic Proteins (BMPs) play a major role in establishing the dorsal-ventral (D-V) axis of most bilaterian animals. In the fruit fly *Drosophila melanogaster* the BMP pathway patterns only the dorsal half of the embryo and acts downstream of the Toll/NF-kB signaling pathway. Outside the insects the Toll/NF-kB -Pathway is primarily used for innate immunity and lacks a function for D-V patterning indicating that the employment of Toll/NF-kB signaling in D-V axis formation is an evolutionary novelty of insects. Recent studies in our lab revealed an important role of the Toll/NF-kB pathway in patterning the D-V axis of the short germ flour beetle *Tribolium castaneum*. To address the question when the transition from a mainly BMP dependent patterning system, to one dominated by Toll/NF-kB signaling, occurred within the insects we analyze both pathways in the parasitic jewel wasp *Nasonia vitripennis*, a representative of the most basal branch of the Holometabolous insects. In addition, *Nasonia* has a *Drosophila*-like, however independently derived, long germ mode of embryogenesis. These characteristics make *Nasonia* an ideal model system with which to understand the evolution of D-V patterning mechanisms.

P-171 The role of Toll signaling for DV axis formation in the milkweed bug, *Oncopeltus fasciatus*

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BMP signaling plays a conserved role for dorsoventral (DV) patterning in most bilateral animals, while in Drosophila embryos DV patterning relies largely on Toll signaling with a limited influence of the BMP pathway. Currently, it is not clear when the Toll signaling was recruited for its function in DV patterning during the arthropod evolution. To gain insights into the evolution of DV patterning in insects we used parental RNAi to study Toll and BMP signaling components in the milkweed bug *Oncopeltus fasciatus*. We show that Toll signaling is indispensible to sustain proper DV patterning in the early embryo. Upon knockdown of Toll signaling, the embryo is dorsalized and the mesoderm anlage abolished. In contrast, the embryo is totally ventralized in the absence of Dpp, the major BMP ligand in *Oncopeltus*. Thus, BMP signaling is required to restrict ventral cell fates. The interplay between BMP and Toll signaling shows that the DV patterning system of *Oncopeltus* heavily relies on BMP and its extracellular modulators while Toll signaling acts only as a polarizing cue.

P-172 The roles of neoblasts on regeneration and reproduction in the annelid, *Aeolosoma viride*

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Some animals can regenerate their lost tissues or organs. During regeneration, both tissue remodeling and organ reformation need cell proliferation and/or differentiation. Neoblasts, one of the somatic stem cells, can proliferate to increase cell number in invertebrates, such as hydra or planarian. Although hydra and planarian are good model animals for regeneration research, their body plans are too primitive to study regeneration of complicated organs. *Aeolosoma viride*, a fresh water annelid with 10-12 segments, a central nerve system, nephridia, and a complete digestive system with a pharynx and an anus, presents great regenerative ability. To identify and trace its neoblasts, three marker genes including Avipiwi, Aviago3, and Avivasa have been cloned, and these genes are highly expressed both in the posterior and in the regenerating tissues of A. viride. Moreover, cell proliferation rate is higher at these regions. Neoblast is highly activated during generation, first 3 days of anterior regeneration, and last 3 days of posterior regeneration. The reproductive ability and regeneration process were significantly inhibited after 90 Gy r-irradiation. And neoblast activity was not detected under this dose

of irradiation. It indicated that neoblast is essential for reproduction and regeneration. Moreover, by RNA interference (RNAi), Avivasa was confirmed to participate in the proliferation ability of the neoblasts. These data strongly suggested that neoblasts in *A. viride* participate in both reproduction and regeneration, which share similar cellular or molecular mechanisms.

P-173 The roles of Zax and Xbap in frog larval head development

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Anuran tadpoles have several novel, unique structures that only exist in the larvae and perish during metamorphosis. The most drastic of these novelties are the rostralia and the very derived organisation of cranial muscles that goes along with these. The rostralia form a crucial part of a novel feeding apparatus and the new arrangement of muscles is necessary for its proper function. The evolutionary success (measured as species number) of frogs in comparison to other recent amphibians is probably associated with his innovation. The cells that make up these novel structures are neural crest-derived, but the evolutionary origin of the rostralia remains unclear and little is known about the molecular basis for such novel structures. We investigate the molecular basis of the formation of the rostralia using functional knockdown of the bagpipe gene Xbap and the bagpipe related homebox gene zampogna (Zax). Using quantitative-PCR, we found a strong correlation between cartilage formation and the expression of Zax in *Xenopus laevis*. Zax-Morpholino injection causes a fatal deformation of the anterior part of the head and leads to missing rostralia. Higher doses cause a total loss of head structures including mouth, eyes and cartilages. Our findings indicate that Zax is essential for the development of the anuran head. Further investigations will include analysing the expression of Xbap and further genes engaged in chondrification. Using whole- mount antibody staining, we will study the effects of Zax-Morpholino injection on muscles attached to the rostralia. This will lead to a better understanding of how new structures can be built and how the diversity of vertebrate head structures evolved.

P-174 Tissue dynamics in the segmenting growth zone of the milkweed bug *Oncopeltus fasciatus*

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Segmentation is a process that requires dynamic expression of different genes in order to gradually produce a sequentially reiterated, well differentiated tissue, from a source of undifferentiated cells or growth zone. While a segmented body plan is a common feature of all arthropods, the process of segmentation in different arthropods demonstrates a diversity of mechanisms, utilising a seemingly common group of orthologous genes. These genes can be active at different times and have varying morphogenetic roles. The main question that arises is what are the implications of the differences in gene activity on morphogenetic processes and cellular behaviour in the newly forming segments. In order to answer this question we describe the basic expression patterns of a number of key developmental genes (delta, even-skipped, engrailed, caudal) during the process of abdominal segmentation in the short-germ hemimetabolous insect, *Oncopeltus fasciatus* (Hemiptera). We correlate these expression patterns with morphological changes in shape and size of the growth zone, and with the pattern of cell di vision therein. We follow these patterns over the process of addition of several abdominal segments and see how they are dependent on the rate of segment addition. A better understanding of the morphological and cellular processes during segmentation can help reconstruct what form the ancestral segmentation mechanism might have had.

P-175 Tooth enameloid in neoselachians: Development, homology, phylogeny

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Chondrichthyans (cartilaginous fish) exhibit a number of interesting features, such as a fully cartilaginous skeleton and continuous tooth replacement that make them valuable organisms to investigate from an evodevo perspective. Their sister group relationship to osteichthyans (that include tetrapods) also offer important outgroup perspectives in the broader scope of gnathostome phylogeny. Since their skeletons are rarely fossilized, isolated teeth are usually the only available material to reconstruct the long evolutive history of this highly successful group. Their teeth are covered by enameloid, an hypermineralized tissue analogous to tetrapod enamel, which precise nature and formation has been the focus of an ongoing debate for over a century. Nevertheless, enameloid microstructure has proved a useful taxonomic tool to differentiate between modern sharks and rays (neoselachians) and their extinct relatives. Recent work has highlighted an unsuspected diversity in the enameloid microstructure in various chondrichthyan lineages, which calls into question both the homology relationships of this tissue among the various members of this group, and the ancestral morphology of the neoselachian and chondrichthyan enameloid. To better understand the origin and modalities of this diversity, supposed to be the result of differences in the composition and structure of the initial extracellular matrix, we investigated early odontogenesis in two

extant models, the small-spotted catshark (Scyliorhinus canicula) and the starry ray (Raja asterias) through classic histological techniques and in situ hybridization. For the later, we focused on the expression pattern of five fibrilar collagens in the small-spotted catshark. The results are expected to bring a better understanding of the precise role of odontogenic cells, ameloblasts and odontoblasts, and to shed new light on the taxonomic value, phylogenetic distribution and homology relationships of the enameloid cover in sharks and rays.

P-176 Transcriptomics of post-anal tail regeneration in the European Amphioxus, *Branchiostoma lanceolatum*

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Amphioxus, the only extant members of the cephalochordata and the basalmost chordates, are important for understanding the evolution and development of chordate characters. This is largely due to their relative simplicity when compared to either vertebrates, which have undergone multiple rounds of whole genome duplication, or urochordates, which have highly divergent adult body plans (Bertrand and Escriva 2011). The European amphioxus *Branchiostoma lanceolatum* is an emerging system model for studying the evolution of regeneration in chordates. While regeneration capacity is poor in many vertebrates, adult amphioxus can reliably regenerate the post-anal tail including segmented musculature, the neural tube and the notochord (Somorjai et al. 2012). All chordates possess a post-anal tail at some stage of development, making it an important comparative character for studying regeneration. As the neural tube and notochord are homologues of the human spine and nucleus pulposus of the intervertebral disc, respectively, gaining insight into how they regenerate in amphioxus has important biomedical implications. We have generated 454 pyrosequenced de novo transcriptomes for *B. lanceolatum* in order to compare the unamputated adult tail with the two-week post-amputation regeneration bud. We have identified several key developmental pathways that pattern the chordate tail during development, including the Wnt and BMP pathways, which are also active in the regenerating tail blastema. Both of these pathways are known to be involved in examples of vertebrate post-anal tail regeneration (Beck et al. 2009). These data will be contrasted to the existing developmental *lanceolatum* transcriptome (Oulion et al. 2012), both in terms of overall gene ontology and presence/absence of individual regeneration-related developmental pathway genes. In addition, quantitative differences in expression of candidate genes will be validated using qPCR both in adult tails and during the regeneration process. This study will provide the first insight into the regeneration transcriptional profile of a complex structure in amphioxus.

P-177 Transformation of skeletal patterns from fins into limbs via a mode change of Turing mechanism

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The skeletal patterns of fish fins and tetrapod limbs are quite different from each other, yet their developmental gene regulations seem highly conserved. Therefore, a slight modification of developmental systems would have been a critical cause of the fin-to-limb transformation. Because a Turing mechanism is increasingly recognized to underlie the developmental systems of skeletal patterning of mouse limbs, we hypothesized that the fin-to-limb transformation can be explained by modification of kinetic parameters of Turing mechanism. To address this hypothesis, we examined pectoral fin development of a catshark, *Scyliorhinus canicula,* and built an *in silico* fin growth model implementing Turing mechanism. We observed that an early chondrogenic marker, Sox9 expression formed periodic spot patterns in *S. canicula* pectoral fin buds. Because mouse limb buds only form continuous stripe patterns, the spot like expression is a specific patterning process of distal radials of *S. canicula* pectoral fins. Furthermore, our *in silico* model showed that modification of parameter values of Turing systems can explain the different dynamics of fin and limb skeletal patterning. Overall, our study gives a clue to understand how similar gene regulatory network create different morphological shapes.

P-178 *Tribolium castaneum* whole embryo culture gives insights into the molecular mechanisms and cell dynamics during body segmentation in arthropods

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In most arthropods segments form sequentially from a posterior growth zone in an anterior to posterior fashion, just like vertebrate somites arise from presomitic mesoderm. It also seems that in both phyla, segmentation relies on the dynamic expression of cyclic genes belonging to the same signaling pathways. However, much of what we know about segmentation in arthropods comes from RNAi-based loss of function analysis, with the corresponding unwanted early effects. Furthermore, it has not been always possible to relate the phenotype obtained with the specific affected part of the process. In

order to exceed these problems we developed a whole embryo culture approach to test different signaling pathways inhibitors/activators during specific time intervals in such a way to determine changes in the segmentation period, elongation rate, wavefront establishment in the growth zone and cellular dynamics. We are already analyzing the effect of SU5402, DAPT and IWP-3, the pharmacological inhibitors of FGF, Notch/Delta and Wnt signaling pathways, respectively, as well as the Wnt signaling activator, LiCl. We found that activation of Wnt pathway affects axis elongation in dissected and bisected embryos, as well as partially disrupts convergent movements toward the ventral midline. On the other hand, DAPT treatment shows embryos with similar germband length and width, when compared with controls. We are now analyzing their segmentation pattern and the expression of the cyclic genes odd- and even-skipped, as well as performing time-lapse cell imaging.

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P-179 Trichohyalin-like proteins have evolutionarily conserved roles in the morphogenesis of skin appendages

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S100 fused-type proteins (SFTPs) such as filaggrin, cornulin and trichohyalin are critical for the development of the skin barrier and skin appendages in mammals. Here we identify, by comparative genomics, SFTPs in sauropsids (birds and reptiles). Orthologs of mammalian cornulin and a trichohyalin-like SFTP termed scaffoldin (SCFN) were characterized in the chicken. As determined by mRNA in situ hybridization and immunohistochemical stainings, both SFTPs were expressed in the periderm of the embryonic epidermis and in the epithelium underneath the forming tips of the claws, at the borders of the papillae of the tongue and in the epithelial sheath around the growing feathers. This expression pattern is similar to that of mammalian trichohyalin, which is present in the nail apparatus, in the filiform papillae of the tongue and the inner root sheath of the hair. These results suggest an evolutionary origin of SFTPs in a common ancestor of mammals and sauropsids and define conserved roles of trichohyalin-like proteins in human and chicken skin. Importantly, our data establish an evolutionary-developmental link between the periderm, a transient embryonic layer of the epidermis, and scaffolding epithelia that support the growth of hard skin appendages in adult amniotes.

P-180 Unexpected function of novel Wnt/Beta-Catenin target genes in Hydra head and foot regeneration

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As in higher animals, also in cnidarians the Wnt/β-Catenin pathway plays a crucial role in developmental and regenerative processes and shows high conservation of structure and function of its signaling components. Wnt/β-Catenin signaling acts in the formation of a head/blastoporal organizer, establishing positional information along the body axis of cnidarian polyps. How this positional information affects cell behavior and to what extent it is evolutionary conserved is unknown. To approach this, a systematic expression profiling of β-catenin target genes was performed. Pharmacologically treated Hydra polyps showed increased or decreased β-catenin levels along the body column. RNA was isolated for quantitative sequencing and compared with wildtype transcripts. Genes with the strongest change in mRNA expression were independently tested via semi-quantitative RT-PCR. Whole mount in situ hybridizations were performed with 10 critical candidates, seven of the genes are so far not described as β-catenin targets. Overall, these genes show activation in the hypostome, in the tentacles, or in an apical gradient in the body column — areas of Wnt/beta-Catenin action. Surprisingly, all target gene candidates show an upregulation in early head and foot regeneration, which provides evidence for direct Wnt/β-Catenin regulation. To uncover direct binding of Tcf/β-catenin to the enhancer regions, CHIP analyses are performed using an α-HydraTcf antibody.

P-181 Using closely related C3 and C4 Flaveria species to define the C4 dicot leaf developmental gradient

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All green plants use photosynthesis to harness the energy of the sun. While most plant species use the ancestral type of photosynthesis called C3, some plants have evolved a more complex version of photosynthesis known as the C4 pathway. C4 photosynthesis tends to be more productive in tropical and sub-tropical environments compared to C3 photosynthesis. Leaves of plants using C4

photosynthesis possess a different anatomy compared with C3 leaves such that vein density is increased and bundle sheath cells are enlarged compared to mesophyll cells. While the underlying biochemical changes are fairly well understood, the development and the genetic regulation of C4 anatomy is yet to be uncovered. Interestingly, C4 photosynthesis has evolved independently at least 60 times across the angiosperms and in many of these lineages the anatomical features are very similar — a striking example of convergent evolution. I am investigating leaf development of four closely related C3 and C4 plants of the genus Flaveria in the Asteraceae to analyse leaf development associated with C4 photosynthesis. Following a detailed morphological study of leaf maturation, I confirmed that young leaves of plants from both photosynthesis types contain gradients in leaf development from tip to base, with the tip possessing the same anatomy as mature leaves and the tissue at the base being undifferentiated. To understand the genetic regulation underlying the differences in C3 and C4 leaf anatomy development, I conducted an extensive RNA sequencing experiment along the leaf maturation gradient that defines the patterns of gene expression associated with leaf development in these C3 and C4 species. Furthermore, comparison of these results with an analogous experiment on leaves of *Gynandropsis gynandra* (formerly *Cleome gynandra*), an independent C4 lineage, and the closest C4 relative of *Arabidopsis* provides insight into the convergence in patterns of gene expression driven by the evolution of this complex trait. These data will be discussed in this context, but also analysed in terms of insights they provide for engineering C4 photosynthesis into C3 crops in order to increase yield.

P-182 VAST-DB: A comparative framework for alternative splicing and gene expression across vertebrate species

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Alternative splicing — the process by which different pairs of splice sites in precursor RNAs are joined to create multiple mRNA variants — represents a major step in the expansion of the proteomic and regulatory complexity of vertebrate genomes, impacting 95% of human multiexonic genes. To obtain a comprehensive picture of the prevalence, regulation and evolutionary conservation of alternative splicing during vertebrate embryonic development and adulthood, we developed a novel transcriptomic analysis pipeline. It integrates various modules that use RNA-seq data, EST/cDNA evidence, gene annotations and evolutionary conservation to identify all existing splice sites, and define all types of alternative splicing in a given species. Furthermore, it uses exon-exon junction mapping RNA-seq reads to accurately quantify the percent of transcript inclusion for each alternative sequence in a given RNA-seq sample. Using this analysis pipeline, we have profiled the different types of alternative splicing, as well as gene expression levels, across dozens of developmental stages, and cell and tissue types in five vertebrate species (human, mouse, chicken, xenopus and zebrafish), as well as key invertebrate outgroups. These data collectively allowed us to identify thousands of AS events that are differentially regulated in a cell-, tissue-, and developmental stage-specific manner, including many that are lineage-restricted and highly conserved across vertebrates. Additionally, the pipeline identified a large set of novel 3-15 nucleotide-long "microexons", which are highly conserved and have intriguing links to vertebrate development and human disease. Information generated from our study will be integrated in a public, web-accessible database, Vertebrate Alternative Splicing and Transcription DataBase (VAST-DB).

P-183 Vent side story

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During early brain development, two organizing centres within the neural plate pattern forebrain, midbrain and hindbrain. One of the organizing centres for mid- and hindbrain is the so-called isthmic organizer or the mid-hindbrain boundary (MHB). Development of MHB is strictly regulated by transcription factors and signalling molecules that form complex gene regulatory network (GRN). These transcription factors (Otx2, Gbx2, Pax2, En2…) and signalling molecules orchestrate expression of Fgf8 and Wnt1, the most studied markers of MHB, into very narrow stripes. In developmental biology textbooks, Vent homeobox transcription factors are associated with establishment of dorso-ventral axis of vertebrate embryos. Recently, Vents have been shown to employ various functions. Intriguingly, these important genes have never been associated with brain development despite their conserved expression profiles in MHB of Xenopus, zebrafish and medaka, suggesting that Vent genes may play a role in development of this extraordinarily important part of the brain. Our results show that medaka Vent expression begins in late gastrula stage. Expression of Vent mRNA seems to be overlapped with Wnt1. In the neurula stage, Vent is expressed in dorsal part of neural tube, similarly to other MHB markers. Vent knock down shows midbrain developmental defects. Concomitantly, Vent overexpression in gastrula stage disrupted expression of Fgf8 and Gbx2 exclusively in the MHB region. The expression of Pax2 and En2 was downregulated. On the other hand the expression of Wnt1 seems to be enhanced, however, the Otx2 expression remains untouched. These changes in expression profiles of

transcription factors and signalling molecules suggest that Vent plays key role in MHB patterning. Our ultimate goal is to establish role of Vent gene in GRN responsible for MHB formation.

P-184 Whence the womanizer? A transcriptomic approach to sex determination in Nasonia

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Nasonia is a genus of small parasitoid wasps. Sex determination in these wasps is particularly interesting, as they are haplodiploid, meaning that fertilized eggs will yield diploid females, whereas unfertilized eggs will yield haploid males. However, it has been clearly demonstrated that sex identity is not directly dependent upon the ploidy of the embryo but rather on levels of the female specific splice form of Nv-transformer, a sex-specific splicing factor in the sex-determination cascade of Nasonia. Since direct genomic imprinting of maternal Nv-tra has been ruled out, recent studies have suggested an unknown factor (termed womanizer (wom)) may be responsible for the zygotic activation of Nv-tra. This factor may be maternally silenced during oogenesis, to ensure maleness in unfertilized eggs, and its mechanism remains unknown. In this study, we set out to analyze the expression and function a set of transcripts that are differentially expressed between male and female Nasonia embryos.

P-185 Wnt pathway is implicated in axial patterning and regeneration in the demosponge *Halisarca dujardini*

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In Eumetazoans, the Wnt pathway is involved in multiple processes during development and regeneration, including symmetry breaking, body and organ patterning, morphogenesis, cell fate determination, proliferation, polarity and migration. All key components of the Wnt pathway are present in sponges, and the pathway is implicated in patterning of body axis during embryonic development and adulthood. Intriguingly, the complexity of the Wnt pathway differs dramatically between previously studied representatives of demosponges (*Amphimedon queenslandica*) and calcisponges (*Sycon ciliatum*), as exemplified by presence of only three Wnt ligands in *Amphimedon* versus 21 in *Sycon*. *Halisarca dujardini* is a demosponge very distantly related to *Amphimedon*, and its development and regeneration are well studied at the morphological level. We have used Illumina technology to sequence transcriptome and generate preliminary draft assembly of the genome of this species. Multiple Wnt pathway components were identified, including ten Wnt and five frizzled genes, in addition to single disheveled and beta-catenin genes. Thus, the complexity of the pathway is intermediate between the previously studied sponges, and appears to be derived from independent gene loss and expansion events. We are now investigating expression of the Wnt pathway components in intact and regenerating adult *Halisarca*. So far we have found that transcripts of at least four Wnt genes are differentially expressed along the body axis, and one is expressed in cells at the edge of wound during regeneration. We expect our results will bring insights into molecular mechanisms of axis patterning and regeneration in demosponges, and into the evolutionary history of the Wnt pathway roles in the metazoans.

P-186 A synchronous patterning model for the development of the vertebrate autopod

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Computer models of vertebrate limb development mostly assume a proximo-distal, tree like growth of the skeletal structures in the forming limb bud. However, such a tree like growth pattern is not present during the development of the autopod. In mice that have been stained for Sox9, limb structures do not begin as differentiated units that grow distally. Instead, there forms a simultaneous pattern of prechondrocyte condensations with interconnections and oval shaped cavities. A clear pattern of longitudinal condensations of the autopod elements becomes visible only during early chondrogenesis. Our approach uses a two-dimensional activator-inhibitor model with graded cell distributions to simulate development of the autopod in the wild type and the results from various polydactyl forms of Maine Coon cats, as well as from human polydactyly. Autopod patterning is simulated as a two-dimensional pattern that operates globally in the domain and not just within a narrow distal zone. The model is able to simulate all five of the phenotypic variations in Maine Coon cats carrying the Hemingway mutation: an extended first toe, a bifurcation of the first toe, additional toes, bifurcations, and free floating toes. We conclude by simulating and interpreting threshold effects in early limb bud chondrogenesis.

P-187 An EvoDevo perspective of primate mirror neuron system through the lens of epigenetics

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The discovery of mirror neurons in primates is one of the major neuroscientific breakthroughs in the last 50 years, among other things for the heuristic potential they represent in cognitive psychology and neurobiology. Thus, understanding

mirror neurons from an evolutionary perspective can highlight some important aspects of the origin of human mind.

After explaining the inadequacy of previous evolutionary accounts, an evo-devo perspective of mirror neurons will be proposed: circuits of mirror neurons, firing during both behaviour observation and execution, could be the result of a process of stabilization of environmental-induced phenotypic traits, in which epigenetic regulation is at the interface between the genetic programming of those neurons and specific social stimuli. From a developmental point of view, epigenetic regulation likely underlies both the origin and the variations of the "basic mirror neuron system" for hand and mouth actions in monkeys. Once shaped the neural connections between sensorial, motor and somatosensory brain areas for the production of facial and arm goal-directed actions, a process of perceptual learning allowed the same areas to be recruited during the perception of both own and others' actions. Epigenetic regulation of preexisting multi genetic loci probably supported these processes. The emergence of mirror neurons has likely brought the individuals to improve their social skills, helping them to recognize actions and intentions of others or to imitate communicative hand gestures and facial expressions. During primate evolution, these social functions might have led to a sort of developmental facilitation of mirror neuron emergence. In particular, behavioural and neurophysiological findings suggest that mirror neurons encoding mouth actions in premotor cortex of macaques and humans may be strongly canalized. The automatic imitative responses of primate newborns to the affiliative gestures of a model and their predictive relation with the development of later cognitive functions suggest a process of genetic assimilation of this trait. Another example of interaction between neural plasticity epigenetically regulated and functions with probably evolutionary value involving mirror neuron systems is the acquisition of tool use in monkeys. In fact, in naïve monkeys, tool use training produces specific mechanisms of neural plasticity in mirror areas of the brain, increasing their human-like cognitive skills, such as planning and imitation. This opens an interesting scenario about the evolution of behaviorally induced brain plasticity, such as that correlated to hand mirror neurons in primates. The manipulation of tools could have constituted a new and challenging environmental niche, leading human ancestors to undergo profound neural plasticity. Thus, changes in mirror neuron-related specific brain areas may have had evolutionary effects via epigenetic phenomena.

P-188 Development of the thalamo-DVR tract in turtles with reference to the evolution of thalamo-telencephalic projection in anmiotes

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In vertebrates, much of the sensory information is transmitted via the thalamo-telencepalic tract. The trajectory of the thalamic axons seems to be highly conserved in the diencephalon of amniotes. However, they take on different terminals among amniote lineage; i.e., in mammals (synapsids), thalamic axons project mainly onto the neocortex, whereas they project onto the dorsal ventricular ridge (DVR) in reptiles and birds (diapsids). To reveal the development and evolution of the thalamo-telencephalic connection in amniotes, we studied the developmental course of thatalmo-DVR tract in reptiles. Neural labeling and gene expression analyses of the axon guidance molecules using the Chinese soft-shelled turtle (*Pelodiscus sinensis*) revealed that the thalamo-DVR connection was formed during the developmental period, and that transcripts of axon guidance molecules were expressed in the diencephalon, similar to the mouse. On the other hand, in the telencephalon, our data indicated the differences in the gene-expression patterns of axon guidance molecules including Slit2, Netrin1 and EphrinA5 between synapsid and diapsid lineages. This result indicates that the local changes in the expression pattern of axon guidance cues may be involved in the diversification of thalamo-telencephalic projection in amniotes.

P-189 Developmental stability and modularity in segmented animals

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The repetition of serially homologous structures along the main body axis is a common form of modularity, found in the body architecture of many multicellular eukaryote taxa. Several studies have explored how natural selection may have promoted the evolution of a modular body organization, and the potential of repetitive body units in taxa diversification. However, comparably less attention has been paid to the possible influence of the developmental features of modularity on the evolvability of this primary morphological trait. Here we present a preliminary essay on a possible relationship between modularity and developmental stability, in the form of a trade-off between the number of body modules and the precision of their phenotypic

expression. This idea has been suggested by the observation that in geophilomorph centipedes, a multi-segmented arthropod clade, most cases of morphological abnormality are recorded in taxa with particularly long and polypodous species. In a sample of eight species, representative of two geophilomorph genera with sizable inter-specific variation in the number of trunk segments, we tested the correlation between different indexes of developmental stability and the number of repetitive body modules, at the level of species and individuals. Developmental stability indexes are calculated using a geometric morphometrics approach, as measures of normalized translational asymmetry for size and shape of segmental structures. These are represented by the spatial configuration of a set of serially homologous setae present on the ventral sclerites of nine contiguous trunk segments. We found no significant correlation between the number of trunk segments and developmental precision at the level of species. Conversely, we found a weak but significant correlation at the level of individuals, which span a range of leg-bearing segments between 43 and 139. Other studies will be necessary to assess the generality of these results, with respects to different segmental features and in a wider taxonomic context.

P-190 Distribution of sea anemone cell types challenges germ layer homology

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Cnidarians (e.g. sea anemones, jellyfish) develop from two germ layers, the outer ectoderm and the inner endoderm, while bilaterian animals (e.g., flies, worms or vertebrates) possess in addition the mesoderm, a third germ layer in between endoderm and ectoderm. It is commonly assumed that the cnidarian endoderm (or "endomesoderm") shares a common evolutionary origin with both the bilaterian endoderm and mesoderm. In order to test this hypothesis, we characterised the localisation, transcription factor profile and embryonic origin of muscle, nutrient storing and digestive cells as representatives for "endodermal" and "mesodermal" cell types in the sea anemone, *Nematostella vectensis*. We found that both the pharynx and the distal parts of the gastric cavity infolds, the septal filaments, are reminiscent of the bilaterian endodermal midgut by the localisation of digestive and insulinergic gland cells within a foxA-expressing region. Strikingly, we could show by using embryonic transplants of transgenically marked tissue that the pharynx and septal filaments derive both from ectoderm in *Nematostella.* The *Nematostella* endoderm, in contrast, harbours muscle cells and nutrient storing cells with transcriptional profiles very reminiscent of bilaterian mesoderm. This allows us to propose a new concept of germ layer evolution where bilaterian endoderm shares no common ancestry with cnidarian endoderm, but with the pharyngeal ectoderm.

P-191 Effects of ROCK inhibitor Y-27632 on *Ephydatia muelleri* development (Porifera, Demospongiae)

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Recent interest in basal lineages of metazoans (Porifera, Ctenophora and Placozoa) has raised fundamental questions concerning the origin and evolution of animals. Since the Rho-Rock pathway has been described as a major signaling pathway in morphogenesis initiation during bilaterian development, studying its implication during sponge development appears now crucial to reveal the ancestral function of this pathway. In the present study, focused on *Ephydatia muelleri* (Porifera, Demospongia), we explored the involvement of this key pathway for the first time in a non-bilaterian species. In the course of gemmule development, ROCK inhibition using Y27632 indices alterations of the edge of gemmule growth leading to outgrowth formation. Moreover, extending treatment (72h) abolishes the establishment of aquiferous system and thus leads to the loss of juvenile body plan. Transcriptomic analyses of treated gemmules by qPCR revealed that both Em-cyclin D (ccnD) and Em-DNA (cytosine-5)-methyltransferase 1 (dnmt1) were over-expressed, thus consistent with a potential cell proliferation. According to the literature, cyclins and dnmt1 are targeted genes of the canonical Wnt pathway to promote cell growth. Therefore, aberrant activation of ccnD and dnmt1 in treated sponges, in addition to outgrowth formation, allowed us to suggest a potential crosstalk between the canonical Wnt pathway and the Rho-Rock signaling. In contrast, expression comparisons of Em-Dishevelled (dvl) and Em-Dishevelled associated activator of morphogenesis (daam) between treated and untreated gemmules showed that the loss of ROCK activity led to the down-regulation of these components. This also suggests that interactions between Wnt pathway and the Rock pathway may exist. To conclude, as it was previously shown in bilaterians, ROCK inhibition during gemmule development improves the key function of this protein in sponges. Contrariwise, its misregulation results in the loss of sponge body plan, and unexpectedly reveals several markers of cancer cells.

P-192 Epithelial morphogenesis during early embryogenesis in *Tribolium castaneum*

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Insects employ a variety of developmental strategies to achieve a relatively stereotypic body plan. We are focusing on the red flour beetle *Tribolium castaneum* that provides an ideal system to study early embryonic morphogenesis and compare it to Drosophila and other insect models. In contrast to the long-germ type of embryogenesis seen in Drosophila, *Tribolium* is a short-germ insect that retains many ancestral characters common to most insects. During early embryogenesis in *Tribolium,* the blastoderm differentiates into an embryonic and an extra embryonic region. This is accompanied by the condensation and folding of the embryonic primordium to form the embryo and the amnion that covers its ventral side, and by the expansion of the extraembryonic serosa that surrounds the entire egg surface. Recently, it has become possible to image live *Tribolium* embryogenesis using transient fluorescence labeling methods and suitable transgenic lines. Considering also the ease of knocking-down genes by RNAi in *Tribolium*, we are using these integrated approaches to study the molecular and cellular basis of embryonic tissue morphogenesis. A major focus will be to understand the cell and tissue mechanics that drive embryo condensation, serosa epiboly, as well as initiation and expansion of amnion folding. We aim to image early *Tribolium* embryogenesis using multi-view Selective Plane Illumination Microscopy and use these recordings to follow nuclear dynamics, cell shape changes and polarized activity of contractile actomyosin networks and junctional components. Our predictions about the forces acting at the *Tribolium* epithelium will be functionally tested by genetic perturbations and laser ablations. This information will be used to generate quantitative models of epithelial morphogenesis in *Tribolium* and other short germ embryos and compare these to the Drosophila paradigm.

P-193 Evolution and constraint in microRNA flanking sequences facilitate their utility in resolving animal phylogeny

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MicroRNAs are a class of conserved 21-23 nucleotide non-coding RNAs that post-transcriptionally regulate gene expression levels in animals. Similar to many protein- coding genes, microRNAs are transcribed inside the nucleus as a long primary transcript before further processing. In the case of microRNAs, processing involves the formation of a hairpin-loop prior to the mature microRNAs in the cytoplasm. Previous studies have suggested that once a novel microRNA is incorporated into the regulatory network of an animal, it is typically retained in the genome during evolution. Given this property, the presence/absence of data of mature microRNAs has been used in recent years as a character to resolve the phylogenetic positions of many animal phyla. Here, we successfully extend the phylogenetic utility of microRNAs by demonstrating the value of their flanking sequences in resolving the phylogeny of animal species. Comparison of all twelve published drosophilid genomes shows that the flanking regions of the hairpin structure are highly conserved. Estimation of phylogeny using the flanking sequences of these drosophilid species reveals an accurate, highly supported tree topology, congruent with the existing evolutionary hypothesis based on concatenated nuclear gene alignments. In contrast, due to an apparent lack of finer-scale resolution in the data, population-level analyses of geographically distant drosophilids fail to resolve evolutionary relationships. This study establishes a new method in estimating animal species relationships at the intra-genus and -family level, which is of wide applicability given the increasing number of available animal genomes. Furthermore, the results suggest that the conserved flanking regions surrounding microRNAs are under strong functional constraint, highlighting an important target for revealing potential *cis*-regulation of microRNAs in animal speciation.

P-194 Evolution and regulation of the chordate ParaHox genes

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The ParaHox genes play an integral role in the anterior-posterior (A-P) patterning of the nervous system and gut of most animals. By comparison between different phyla, as well as with their evolutionary sisters, the Hox genes, it has become clear that both the Hox and ParaHox genes display remarkably similar regulatory phenomena. Evidence now points to the presence of clusters of Hox and ParaHox genes displaying colinearity in the last common ancestor of protostomes and deuterostomes, with the order of the genes in the cluster corresponding to the order of their activation along the embryo. Whilst these phenomena are well studied in the vertebrate Hox system, the ParaHox cluster of the invertebrate chordates offers a much simpler system to investigate the evolution and regulation of these regulatory processes as it possesses only three genes, compared to the much larger Hox cluster. The invertebrate chordates offer

two different "systems", of intact and dispersed clusters, in which to study ParaHox regulation with the additional benefit that these invertebrate chordate lineages evolved before the whole genome duplication events that occurred at the origin of the vertebrates. Using the cephalochordate amphioxus and the tunicate *Ciona intestinalis* we show that the chordate ParaHox genes are regulated by key developmental pathways involved in axis specification, providing evidence for ancestral mechanisms regulating both the Hox and ParaHox genes at the base of the chordates.

P-195 Evolution of Prdm genes in animals: Insights from comparative genomics and gene expression studies

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Prdm genes encode transcription factors with a PRDI-BF1 and RIZ homology (PR) domain and a variable number of zinc finger motifs. These genes show a wide variety of functions. In particular, several Prdm genes, such as Prdm1 (also known as Blimp1) and Prdm14, have been shown to have important roles in somatic pluripotent stem cells and in primordial germ cells. Other genes, such as Prdm8, Prdm12, and Prdm13, are expressed in specific neural populations and are required for the proper development of these neural cells. Whereas the functions of Prdm genes have been carefully studied in some vertebrates, especially mouse, little is known about the evolution of this gene family. We have searched for Prdm genes in the fully sequenced genomes of 91 different animal species representative of all the main animal lineages. We identified a total number of more than 900 Prdm genes in these species, the number of Prdm genes per species ranging from 2 to 19 depending on the species. To better understand how the Prdm gene family has evolved in metazoans, we performed phylogenetic analyses using the large set of Prdm genes we have identified. These analyses allowed to define 14 different subfamilies of Prdm genes and to establish that 11 of them are ancestral to bilaterian animals. Detailed analysis allowed to define the gene duplication and gene loss events that occurred in the different animal lineages. By studying a large number of non-animal genomes, we also defined the most likely evolutionary origin of this gene family. To get insight into the evolution of the functions of these genes in bilaterian animals, we cloned the full set of Prdm genes from the emerging model species, the annelid *Platynereis dumerilii*, a slow-evolving species that is distantly related to both vertebrates and arthropods. Expression patterns of the cloned genes will be reported. Together, our data provide new insights in the evolution of this important family of transcription factors.

P-196 Evolutionary changes in proneural gene expression: Atonal and ASH in *Daphnia magna*

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In insects the large number of sense organs can be subdivided into groups based on their morphology and corresponding function. In *Drosophila melanogaster* sense organ development requires proneural genes for the selection of sensory organ progenitors (SOPs), but also for their subtype identity. For example, members of the Achaete-Scute family are required for external mechanosensory organ development, while members of the Atonal family specify chordotonal organs and a subset of olfactory sense organs among others. Thus, the expression domains of these genes do not overlap in *D. melanogaster*. We investigate the morphological and molecular development of sensory organs in *Daphnia magna*. Surprisingly, we found that ASH and atonal are expressed in an overlapping pattern in several areas of the peripheral nervous system. Examples are the distal parts of the antennae, the mandibles and the thoracic appendages, as well as the posterior margin of the proctodeum. Since it is not known which sense organs are generated in these areas, we are analyzing the structure of larval sense organs and trace their origin back to embryonic stages. The partial co-expression of ASH and atonal in *D. magna* indicate that either the molecular mechanisms in sense organ determination have changed during crustacean evolution or that sense organs are formed that show characteristics distinct from insect sense organs. Therefore, we generated a transgenic *D. melanogaster* line, which carries the *Daphnia* atonal gene in order to perform over-expression and rescue experiments. Interestingly, *D. magna* atonal does not rescue the atonal-mutant phenotype, but is able to restore external mechanosensory organ development in the absence of achaete-scute function.

P-197 Expression of acetylcholinesterase during development and regeneration of the *Octopus vulgaris* arm: Indications of a "non-classical" role

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The Octopus arm crown consists of eight amenable and flexible arms, composed of a three-dimensional network of longitudinally, transversely, and obliquely arranged muscle fibers and connective tissue. An axial nerve cord and a dense network of peripheral nerve fibers regulate the motor control of this so-called muscular hydrostat

and allow the arm to perform complex and precise movements. In addition to its interesting physiological capabilities, the Octopus arm is capable of regeneration and, therefore, provides a great model for comparing muscle formation during development and adult regeneration in an invertebrate organism. In this study we examined the role of Acetylcholinesterase (AChE) during embryonic development and adult regeneration of muscular and neural tissue in the cephalopod mollusk, *Octopus vulgaris*. AChE is a multifunctional glycoprotein, which in addition to its classical-cholinergic function is involved in non-cholinergic mechanisms important for embryogenesis and regeneration in both vertebrates and invertebrates. These include embryonic neurite extension, synaptogenesis, cell adhesion, and muscle formation. We cloned an *O. vulgaris* - AChE homologue, which we identified as an AChE H (hydrophobic) variant and studied its expression pattern during the processes of arm development and regeneration by in situ hybridization and RT-PCR. AChE was mostly localized in seemingly undifferentiated, mesenchymal, and highly proliferating tissue during the early stages of arm embryogenesis and adult morphogenesis. In later stages, at the establishment of an adult-like structure of the arm, AChE expression was mostly identified in typical neuronal (cholinergic) sites. These included the axial nerve cord, longitudinal muscle fibers, and the neuromuscular components of the arm's sucker. Our results suggested a non-classical role of *O. vulgaris* - AChE during early embryonic development that may be conserved throughout the animal's lifespan and reactivated upon injury. In an ongoing study we are attempting to understand possible non-classical functions of *O. vulgaris* AChE during embryonic arm development, by interfering with both the catalytic and the peripheral non-catalytic site of the enzyme using pharmacological inhibitors. The results of this study will give us a more detailed insight into the significance of AChE during formation of the embryonic, as well as regeneration of the adult *O. vulgaris* arm.

P-198 Hox genes of the hagfish and the deep roots of vertebrate genomic evolution: Insights from the embryonic transcriptome of the hagfish

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It is now clear that vertebrates experienced whole-genome duplications (WGD) at their evolutionary origin, but the timing and number of rounds of these duplications in the different vertebrate groups remain unclear. While the two rounds of WGDs (2R) shared by gnathostomes (jawed vertebrates) are very well characterized and studied, it remains unknown whether these 2R-WGDs are also shared by cyclostomes (jawless vertebrates: hagfish and lampreys), or whether cyclostomes underwent (additional) independent WGD round(s). The recent analyses on two lampreys' genomes do not clarify the genomic history of vertebrates: while the marine lamprey genome supports that the 2R-WGD events were shared by the common ancestor of all vertebrates, the more recent genome of the Japanese lamprey does not, and moreover seems to support a 3R-WGD event in the lamprey lineage. In order to definitely settle the question about the early genomic evolution of vertebrates, genomic analyses from the hagfish, the sister group of the lampreys, are needed. Here, we present a preliminary analysis of an RNA-seq of embryonic samples of the Japanese inshore hagfish, *Eptatretus burgeri*, a rare specimen whose embryos are extremely difficult to obtain either in captivity or in natural conditions. We have systematically screened the hagfish transcriptome for Hox genes, and a hagfish BAC library for Hox clusters, since the number of Hox genes and clusters generally reflect the number of WGD events in vertebrates (i.e., four Hox clusters result from 2R-WGD events). In our preliminary analyses, we have found a total of 42 Hox genes, with up to 7 Hox4 paralogues, indicative of the presence of up to 7 putative Hox clusters, supporting a lamprey 3R-WGD in concordance with the Japanese lamprey genome. Hagfish and lampreys also share the lost of all Hox12 paralogues and the presence of a single Hox14 gene, indicating that this 3R-WGD event might have happened in the last common ancestor of cyclostomes, and not independently in each lamprey and hagfish lineages. Our results drastically change the previously depicted scenario of the vertebrate genome evolution and might explain the big morphological disparities between cyclostomes and gnathostomes.

P-199 Insights into arthropod hormone evolution by sequencing two non-insect arthropods: S shrimp and a millipede

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The phylum Arthropoda contains the largest number of living species of animals on Earth. However, most of our understanding of their genomes, developmental patterns and endocrine systems — especially the mechanism of hormonal regulation — mainly comes from the Insecta. Recently we have sequenced the genomes of a new shrimp model (*Neocaridina denticulata,* Crustacea) and a millipede (*Trigoniulus corallinus,* Myriapoda). Here we found the majority of

known insect hormonal pathway genes and their regulators in these non-insect arthropod genomes, including several components not previously reported in the crustaceans, myriapods and chelicerates. The identification of the juvenile hormone and ecdysteroid biosynthetic pathway genes in these non-insect arthropods provide new insights into the understanding of the endocrine system and evolution in arthropods.

P-200 Involvement of Slit-Robo signaling in the development of the posterior commisure and concomitant swimming behavior in *Xenopus laevis*

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The vertebrate brain has specific neuronal tracts called the "early axonal scaffold" that appear during its early development. These first tracts include the longitudinal and commissural axonal bundles such as medial longitudinal fascicle or posterior commissure (pc), which are highly conserved in gnathostomes and also in agnathans. Since these early tracts appear to guide the later-developing neurons, they are thought to provide the basic framework of the vertebrate brain. In this study, to reveal the developmental mechanism underlying the formation of these early tracts, we inhibited axon guidance molecules including Slit2 ligand and Robo2 receptor. We demonstrated that inhibition of Slit2 and Robo2 in *Xenopus laevis* larvae resulted in the abnormal morphology of the pc, a kind of early axonal scaffold. Furthermore, we identified abnormal swimming in Slit2- and Robo2-morpholino-injected larvae. Thus, Slit-Robo interaction appears to be involved not only in the formation of pc but also in the establishment of functional brain elements that triggers a swimming behavior. Together with the fact that the pc has been highly conserved in vertebrate brain evolution, it is suggested that in vertebrates, Slit-Robo signaling will be contributed to the construction of pc, which is a prerequisite for the subsequent development of complex neuronal circuits and functions.

P-201 Manipulation of metamorphic development in sea urchins by morpholino microinjection into late stage larvae

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Sea urchins have been used as experimental organisms for developmental biology for over a century. Their embryos and larvae are transparent, can be reared in large quantities and can be easily manipulated in the lab, both morphologically and genetically. Still, as in many other marine invertebrate groups, understanding the development of juvenile tissues has lagged far behind that of embryos. The reasons for this are not due to lack of interest, but rather, in part, to a lack of experimental approaches to manipulate development. Here we report on the validation of a technique for injecting compounds into juvenile rudiments of the purple sea urchin, *Strongylocentrotus purpuratus*. As a proof of concept for using this technique, we injected vivo morpholinio's (vMOs) designed to knock down P58b and P16, into the juvenile rudiment of sea urchin larvae. These two proteins have been previously shown to be involved in the elongation of *S. purpuratus* larval skeleton. Rudiments injected with these vMOs showed a delay in the growth of some skeletal elements relative to several controls. These data provide the first evidence that vMOs, which are designed to cross cell membrane, can be used to transiently manipulate gene function in late developmental stages. We therefore propose that injection of vMOs into juvenile rudiments, as shown here, is a viable approach to testing hypotheses about gene function during development.

P-202 MicroCT based analysis of chemically perturbed axis formation

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Vertebrate development is regulated via various highly interconnected networks, which are commonly studied by gene knockout and mutation experiments. In this study, by contrast, we analysed chemically induced wavy notochord phenotypes for this purpose. Because this malformation can result from a number of different stimuli, we can use it to study various developmental processes involved, such as cell adhesion, cell migration, cell growth regulation, and matrix metabolism. By treating with PTU to induce a wavy notochord and associated phenotypes, we could also observe an fgf8 mutant-like phenotype — independent from the notochord phenotype — that could be partially rescued with simultaneous retinoic acid treatment. We wanted to draw inferences about which morphogenetic processes are targeted and about their signalling cascade structure and regulatory interplay. Therefore we performed FGF receptor inhibition using SU5402 in order to reveal the hierarchy of the regulatory competence of retinoic acid and fgf8 signalling — which is crucial for the correct setup of brain asymmetry and patterning. The use of high-resolution x-ray microtomographic imaging (microCT) to analyse developing and perturbed morphologies provides unrestricted possibilities for visualisation, as well as quantitative information about deviations from normal development. The volume images can be used

to measure, among other features, normal and abnormal asymmetry in the brain region and differing lengths of notochord and other body regions at critical stages. This work also lays the foundation for planned experiments on the roles of these morphogenetic networks in regeneration.

P-203 Multiple functions of Zerknüllt-2 during early patterning of the short germ beetle Tribolium

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The Drosophila morphogen Bicoid (BCD) is a homeobox protein that not only functions as transcriptional activator but in addition can bind to caudal (cad) mRNA and repress its translation. While posterior expression of CAD is conserved in insects, nematodes and vertebrates, BCD is a unique feature of higher Diptera. Previously, we have shown that in the short germ beetle *Tribolium*, Zerknüllt-2 (ZEN-2) and MEX-3 functionally substitute for BCD in cad translational repression. To further elucidate the functions of ZEN-2, we raised antibodies and generated transgenic heat-shock misexpression lines. Upon heat-shock mediated ZEN-2 misexpression, we observed severe patterning defects. ZEN-2 heat-shock larvae are depleted of most head and thoracic segments and in addition, growth-zone derived segments are reduced in number. We also observed homeotic transformation of abdominal toward thoracic fate. While ZEN-2 misexpression apparently does not effect early CAD distribution, our results indicate a function for ZEN-2 in early patterning independent of CAD translation regulation. In *Tribolium*, ZEN-2 not only is required for repression of CAD translation in the presumptive serosa, but also for the subsequent fusion of amnion and serosa and thus, for dorsal closure. We now provide evidence that ZEN-2 likely fulfills an additional function in early anterior-posterior axis formation. Since both, bicoid and zen are thought to have originated through a duplication of an ancestral Hox3 gene, our data suggest that the function in anterior patterning of Hox3/Zen-like genes preceded the evolution of bicoid.

P-204 New perspectives of study of nervous system formation of Galathowenia oculata (Oweniidae, Annelida)

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Galathowenia oculata is a member of the peculiar tubeworms group Oweniidae. This is a small group of 55 benthic species worldwide-spread from the continental slope to abyssal depths, mostly at soft sediments like mud or sand which particles worms use for feeding and tube construction. The phylogenetic position of the group has been changed many times. Previously they were regarded as a member of primitive basal "Archiannelida", then they were referred to sedentary filter-feeding Sabellidae together with derived "Siboglinidae", then were grouped only with "Siboglinidae", then referred to spionid polychaetes Apistobranchidae, then placed as basal polychaete and recently again they were considered again as a basal group of Annelida. In spite of their evident polychaete-like appearance oweniids do not have typical spiralian embryogenesis; there is gastrulation via invagination like in Deuterostome and Lophophorates, deuterostome mouth-formation, unique mitraria larvae, catastrophic metamorphosis like in Nemertines, Phoronida, Echinodermata and "polychaete" Polygordiidae, monociliated epidermis like in Phoronida, Brachiopoda, Echinodermata, Hemichordata and also Gnathostomilida, Gastrotricha, "polychaete" Magelona, deuterostome-like nephridium, intraepidermal nervous system and muscle regulation through basal lamina like in Echinodermata, Hemichordata and Phoronida, absence of internal dissepiments etc. Thus, combination of the results of phylogenetic analysis, peculiar morphology and unique development pattern can be the reason to speculate that Oweniidae are really basal representatives among Annelida. Recently, the primitive organization of the nervous system of *G. oculata* was shown by the author and colleagues: absence of brain as well as any other ganglia, massive nervous plexus in body wall and numerous longitudinal nerve cords with the main ventral one. It is uncertain if the condition of nervous system is determined by catastrophic metamorphosis, either by the ecology of burrowing animal without numerous appendages and sensory structures, or by basal position within Annelida. In this regard, *G. oculata* is an intriguing model object for evo-devo experiments to understand mechanism of nervous system formation, including such questions like absence of ganglia, plexus development and why the nervous system retains in epidermis. This presentation is necessary for discussion and planning of a new project.

P-205 Origin of FGF signalling

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Complex metazoan bodies require cell-to-cell communication for development, a process partly achieved by signalling molecules binding to specific receptors. Relatively few signalling pathways have been recruited during evolution to build multicellular animals from a unicellular zygote. Fibroblast Growth Factors (FGFs) bind to receptors in the Receptor Thymidine Kinase family, but the origin of the eumetazoan FGF gene family has remained a mystery. We were able to show that extant bona fide FGFs probably originated from proteins

bearing an FGF-like domain shared with their common ancestors, the unicellular choanoflagellates. We found orthologous genes closely related to FGF in choanoflagellates as well as in many metazoan phyla such as sponges, acoels, non-vertebrate deuterostomes or protostomes. We also show that these genes have a common evolutionary history with Retinitis Pigmentosa 1 (RP1). Even if some metazoan signalling pathways emerged long before multicellularity, we show that FGFs and their receptors were innovated in a eumetazoan ancestor.

P-206 Patterns of sexual selection on cranial shape in natural populations: Relative eigenvalue approach

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Complex morphological structures such as the mammalian cranium are subject to various kinds of natural selection that affect their size and shape. Relative eigenvalue analysis enables the identification of cranial traits that are affected by selective pressure, whether divergent or stabilizing. By inspecting the changes in cranial shape between roe deer sexes, the effect of sexual selection on measured cranial traits could be investigated. Cranial shape was quantified by selecting 16 landmarks on one side of ventral projection, from a set of digital photos (90 males and 74 females). Euclidean distances were extracted from all possible pairs of landmarks and a smaller subset was selected through hierarchical cluster analysis based on the correlation distance matrix. A total of 14 distances were selected as cluster group medoids. Separate covariance matrices were calculated from selected distances for males and females, while between-sexes (B) and pooled phenotypic within-sexes (P) covariance matrices were extracted as SSCP matrices from a multivariate model with sex as a grouping variable. Common covariance matrix (P-1B) eigenvalues and eigenvectors (relative eigenvalues-RE, eigenvectors-RV) were calculated and the average RE was used as a scaling factor of B to P. All covariance matrices were then compared using a metric distance function based on RE, and their ordination was explored with principal coordinate analysis. B and P matrices were significantly different, while males and females cluster around the P matrix in the ordination space. Higher positive or negative loadings of the selected distances on the first and the last RVs were interpreted as possibly being under directional or divergent selective pressure, while the ones with loadings close to zero were considered selectively neutral. Results suggest that sexual selection for cranial shape influenced the relationship between lengths of the anterior (premaxillary-palatine) and posterior cranium (palatine-basioccipital) as well as mid-cranial width from basisphenoid to the outer edge of the zygomatic arch. Mid-cranial width is expected to be higher in males, providing better support for the antlers, while antero-posterior length differences may indicate contrasting foraging specializations or different patterns of intra-sexual competition.

P-207 Plant surface texture: Investigating R2R3 MYB subgroup 9 gene function in Marchantia and Nicotiana

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The subgroup 9 gene family of R2R3 MYB transcription factors is an ancient gene lineage that arose prior to the origin of the land plants. They have been functionally characterised only within the flowering plants, where they regulate diverse epidermal structures including petal epidermal cell outgrowths, trichome development, and the initiation and elongation of cotton fibres. Broadly, my PhD seeks to understand the ancestral function of these genes, and their subsequent diversification in the mediation of epidermal structures across different land plant lineages. I will present data on the function of subgroup 9 homologs in the early land plant model *Marchantia polymorpha*, using semi-quantitative PCR, overexpression and amiRNA knockdown. I will also explore the role of subgroup 9 genes in floral evolution in Nicotiana, where I have used sister species to determine if changes to subgroup 9 gene sequence or expression are associated with shifts in petal cell shape.

P-208 Plasticity of hominoid developmental patterns in response to habitat exploitation: Implications for hominin life history evolution

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Studying life history variation in hominoids is hampered by the challenges of studying these animals in the wild and by their long life spans. Yet, an understanding of the development of great apes underpins interpretations of the evolution of hominin life histories, i.e., when and by which mechanisms our extended growth periods evolved. Here we take a cross-sectional approach that combines biochemical and morphological analyses and uses a well-documented Museum collection. Stable isotope analysis on hair was employed to determine how gorillas and chimpanzees from Central Cameroon partitioned their habitat. Results showed complete separation in d 13 C, but not d 15 N, when gorillas are sympatric with chimpanzees, indicating that the former occupy more densely canopied forests. To assess the biological consequences of such habitat partitioning, an ontogenetic series of 35 gorilla and 44 chimpanzee skulls was analysed for patterns of tooth mineralisation/eruption and brain size increase. Evidently, gorillas

develop more slowly than chimpanzees such that only about 87% of adult brain size is attained by the time first permanent molars come into occlusion (93% in chimpanzees). Even when M1s are already in full functional occlusion, gorilla brains lag behind those of chimpanzee (91% versus 99%). The findings are consistent with the "risk aversion hypothesis" for frugivorous species, and with life history theory that predicts delayed development when non-density dependent mortality is low, i.e., in closed habitats. This highlights the plasticity of hominoid life histories and cautions against simplifications when interpreting early hominin life histories. Furthermore, they highlight the importance of dietary ecology and habitat preference for life histories.

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P-209 Quantifying nature's appearance: Combining high-resolution, fully coloured 3D reconstruction and mathematical tools to analyse skin patterns in corn snakes (*Pantherophis guttatus*)

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An expanding number of studies investigate the biophysical mechanisms generating intra- and inter-specific complexity and diversity of morphological traits. However, classification of morphologic characters is often limited to qualitative descriptions or simple quantitative analysis. To address this issue, we have combined state-of-the-art robotics, high-resolution (36 Megapixel) digital cameras and 3D reconstruction Multi-view Stereo and Shape from Shading algorithms to build a scanning system capturing geometry details down to 50 microns and colour texture details down to 20 microns. This system is (1) greatly flexible, allowing a scanning range that extends from above the meter (e.g., a full corn snake) to sub-millimiter details (e.g., within the scales of a reptile); (2) highly repeatable, making it suitable for a systematic approach; (3) fast, with scanning times below 5 minutes, a critical feature when animals under anaesthesia are used. These fully-coloured 3D virtual mesh models are accurate mathematical representations of the real animal and its external morphological traits, and are therefore suitable for the application of mathematical tools. This allows for the systematic and rigorous quantification of features such as colour variations and gradients, repetitive patterns, shapes, areas, or correlation among specific traits. By using the colour patterns of corn snakes (*Pantherophis guttatus*) as an example, we argue that this approach has practical importance for innovative EvoDevo analyses of phenotypes in 3D.

P-210 Stem cell dynamics in the hydrozoan *Clytia hemisphaerica*

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In hydrozoan cnidarians somatic cell types such as neural cells, nematocytes, gland cells, but also gametes, form multipotent stem cells called i-cells (interstitial cells), as has been well documented in Hydra polyps. Much less is known about the formation of i-cells and their derivatives during larval embryonic development. In the hydrozoan experimental model *Clytia hemisphaerica* i-cells have been described morphologically during development to appear at early larval stages and to be localized in the central endodermal region. During embryogenesis the expression of several genes, considered to be stem cell markers and/or germ plasm components such as Nanos1, Nanos2, Piwi, Vasa, and PL10, has been detected in cells derived from a "germ plasm"-containing region at the animal pole of the egg, which in the larva appear to be inherited by a population of cells in the endodermal region putatively equated with i-cells. In this study we provide a detailed description of i-cells and their derivatives during embryogenesis and larva development in Clytia, using a set of molecular markers identified from published work in Hydra, in Clytia and from our group's recent transcriptomics data (unpublished). By systematic in situ hybridization we built up a spatial and temporal map of the distribution of i-cells and their derivatives during Clytia larval development. This characterization opens the way to investigate the signals regulating i-cell formation and commitment to different fates through functional approaches. We are currently testing the roles of Wnt signalling in i-cell formation and differentiation, by injection of morpholino oligonucleotide against Wnt3 and use of pharmacological inhibitors. This study increases current understanding of stem cell behavior in early branching metazoans, and should contibute to deciphering the ancestral roles of Wnt signaling in stem cell proliferation and fate determination.

P-211 Symmetrically and asymmetrically substituted phthalocynanines and toxic effects on Drosophila melanogaster

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The chemical structure of phthalocyanines can be modified by the introduction of substituents in the molecule as well as by the coordination of different metal ions with the central nitrogen atoms. In

the proposed projects, our goal is to show that new phthalocyanines with different functional groups have different photophysical and photobiological effects and that these molecules have potential to be effective on a living organism. To that end, we are planning to synthesize phthalocyanine analogs. Then, toxic effects on these molecules on *Drosophila melanogaster* will be determined. For this purpose, the changes in *D. melanogaster* will be examined as morphological, fertility, and sex ratio.

P-212 The evolution and development of petal spots in the Angiosperms

Mellers, Greg (University of Cambridge, GBR); Glover, Beverley (University of Cambridge, GBR)

Angiosperms are the most diverse division of extant land plants, occurring in almost every environment on Earth, with estimates of around 260,000 to 420,000 species. A key element in the formation of such a speciose group is thought to be their intimate co-evolution with pollinator species. Typically such pollination syndromes are gross features such as corolla colouration or floral scent. However, it is becoming increasingly apparent that finer scale features may also attract wild pollinator species. One such characteristic is the appearance of petal spots on the corolla of some species that have previously been shown to increase reproductive success. It is hoped that formation of this feature in a heterologous crop system may confer greater pollinator interactivity and thus yield. Our study aims to investigate how petal spots develop in the model daisy species *Gorteria diffusa*. Previous work has found there to be multiple "morphotypes" of the species that represent a series of natural mutations fixed within discrete populations. These allow for comparative analyses to be undertaken with the intention of elucidating the molecular regulation of spot generation. Some evidence suggests a role for MYB genes in the spot formation process hence comparative expression analyses (ΔΔCt qPCR) and heterologous expression studies are being undertaken. Furthermore, multiple forms of microscopy (Light, SEM, TEM) are being employed to further characterise the morphology of the spot in the hopes of identifying other candidate regulatory genes. Restriction site Associated DNA (RAD) sequencing is also being employed to acquire high SNP coverage in an attempt to understand the relationship between the aforementioned morphotypes. It is hoped that through these diverse techniques a hypothetical model for spot formation may be found and subsequently perturbed for validation.

P-213 The evolution of a metamorphic life history in the phylum Nemertea

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The life cycle of the pilidiophoran worms is maximally indirect and culminates in catastrophic metamorphosis. They begin as planktonic pilidium larvae that feed and swim using a ciliated band. Over the course of weeks, a juvenile worm arises inside the larva from the fusion of isolated rudiments called imaginal discs. Once the juvenile is formed, it escapes the pilidium and devours the larval body. Remarkably, this type of life history evolved within the nemerteans; the basal members of the phylum and the sister clade to the pilidiophorans (the hoplonemerteans) have a more direct development. The larval and juvenile body plans in pilidial development share little in common, and so it is possible that the underlying patterning mechanisms are also different. Furthermore, the diversity in orientation of juvenile antero-posterior (AP) axis with respect to the larval AP axis within the Pilidiophora hints at uncoupling between the larval and juvenile AP patterning. To gain insight into the morphogenesis of the larval and juvenile bodies, we examined developmental mechanisms in the species *Micrura alaskensis* by studying expression of conserved AP patterning genes, including Hox. We found that Hox genes do not pattern the pilidial larval body. Instead, their expression is limited to the imaginal discs that give rise to the juvenile trunk. Thus, at least one major AP patterning mechanism is not shared between the two life history stages. This may be key to understanding how pilidiophoran development evolved and diversified — with some degree of mechanistic separation that may have allowed for independent evolution across stages. We also examined AP patterning genes in larvae of the hoplonemertean *Pantinonemertes californiensis*. These larvae overtly resemble juveniles and lack a distinct metamorphosis, but possess features that are reminiscent of indirect-developers, such as a transitory larval epidermis and a set of epidermal invaginations that invites parallels to the invaginated juvenile rudiments in the pilidium, i.e., the imaginal discs. Our results from the expression of Hox and Six3/6 genes suggest the homology between these invaginations and pilidial imaginal discs. We discuss the implications of these findings for the evolution of pilidiophoran development.

P-214 The evolution of evolvability

Altenberg, Lee (The KLI Institute, Klosterneuburg, AUT)

The phrase „evolution of evolvability'', coined by Dawkins (1988), was adopted by several researchers to better summarize a theoretical subject that originated with Riedl (1975) and Conrad (1977). I will

use „evolvability“ to mean the upper tail of the distribution of fitness effects of genetic variation. While many assert that the evolution of evolvability is „controversial“, the evolution of the upper tail of fitness distributions actually goes back to Fisher (1930). „Evolvability“, when understood as the upper tail of offspring fitness distributions, is clearly not a „population-level feature“ requiring group or lineage selection to evolve, but is a property of individuals. The upper tail will evolve when it co-varies with individual fitnesses, as described by the Price (1970) equation. In the usual model of a population evolving toward an adaptive peak, as in Fisher's geometric model, the covariance will be negative, and the upper fitness tail will shrink. More challenging is to identify evolutionary mechanisms that systematically cause the upper fitness tail to be maintained or to expand. My contribution is to theoretically tie the upper tail of fitness distributions together with several disparate phenomena: the modularity and complexity of the genotype-phenotype map, gene origin dynamics, the relationship of gene duplication effects to allelic variation, and the consequences of iterative gene duplication over macroevolutionary time. These relationships are still not widely understood, so they will be the focus of my talk.

P-215 The evolution of the vertebrate stomach and the paradox of loss

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The vertebrate stomach represents a unique anatomical innovation, characterized by the presence of acid and aspartic protease secreting glands. However, the presence of gastric glands in vertebrate species is not universal. The French zoologist Cuvier first noted that some teleost species lacked a stomach in the 19th century. Similarly, Chimaeriforms, Dipnoids and Monotremas also lack acid secretion and a gastric cellular phenotype. Here we investigate the specific contribution of gene loss to the widespread distribution of the agastric condition. We establish that the stomach loss correlates with the persistent and complete absence of the gastric gene kit in the analysed species. In gastric lineages, we find also that the pepsinogen gene complement varies significantly with events of pseudogenization identified in various lineages. This variable repertoire likely reflects dietary driven episodes of gene family expansion and contraction. The apparent paradox suggested by the successful retention of stomach and stomach-less species is discussed.

P-216 The expression of Epiregulin in mandibular deciduous molar development of the miniature pig

Fan, Zhipeng (Capital Medical University School of Stomatology, Beijing, CHN)

Purpose: To investigate the expression of epidermal growth factor, Epiregulin (EREG) in the tooth development of the miniature pig.

Methods and Materials: The staged miniature pig embryos at embryonic day (E) 40, 50 and 60 days of gestational age were obtained respectively. Dissected mandibles from staged specimens for histological processing were collected and fixed in 4% neutral paraformaldehyde and decalcified with 10% EDTA or Morse's solution. Demineralized specimens from the free right or left mandibles of each stage were processed for routine dehydrating, clearing, and embedding in paraffin. Serial sections in sagittal or frontal plane at 5 μm thicknesses were prepared. Sections were stained with haematoxylin and eosin (H&E) or immunohistochemistry and examined using light microscopy.
Results: The results of HE staining showed that tooth germ of E40, E50, E60 was in the bud stage, the cap stage and bell stage respectively. The results of immunohistochemistry staining showed that the EREG was positive expressed in the dentin layer, odontoblasts and ameloblasts in late bell stage of mandibular molars tooth.
Conclusions: The result that EREG expression indicated that EREG may play a role in the development of tooth. The significantly increased EREG expression in late bell stage suggested that EREG might involve in the function changes from promoting cell proliferation to cell differentiation.

P-217 The first zebrafish neural crest in vitro model and its application to the study of retinoic acid

Kinikoglu, Beste (Acibadem University, Istanbul, TUR); Kong, Yawei (Harvard Medical School, Boston, MA, USA); Liao, Eric C. (Harvard Medical School, Boston, MA, USA)

Neural crest is a unique, multipotent and migratory cell population associated with vertebrate development. Defects in neural crest development result in a wide range of malformations such as cleft lip and palate, and diseases such as melanoma. We have isolated these unique cells from zebrafish embryos and established reproducible functional neural crest cell behavior assays, in terms of cell proliferation, migration and differentiation. The cells were isolated from transgenic sox10:egfp embryos using fluorescence activated cell sorting and expressed major markers of neural crest cells such as hnk1, p75, pax3, sox9a and dlx2a, and also the markers of pluripotency such as c-myc and klf4. We showed that the cultured neural crest cells can be differentiated into multiple neural crest

lineages, contributing to neurons, glial cells, smooth muscle cells, melanocytes, and chondrocytes. We applied our neural crest cell in vitro model to study the effect of retinoic acid, a vitamin A derivative endogenously synthesized in all vertebrates, on neural crest cell development. We showed that retinoic acid had a profound effect on neural crest cell morphology and differentiation, significantly inhibited proliferation, and enhanced cell migration. Our data implicate neural crest cells as a target cell population for retinoic acid, and strongly suggest that retinoic acid plays multiple critical roles during embryonic development. We hope that our novel neural crest in vitro system will be useful to gain mechanistic understanding of neural crest development and for cell-based, high-throughput drug screening applications.

P-218 The function of Oct4 homologues in the evolution of epiblast versus germ cell potency: Relevance to embryonic stem cell self-renewal and induced pluripotency

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Oct4 or Pou5f1 is a master regulator of pluripotency and differentiation in both embryonic stem cells (ESCs) and during embryonic development. It has conserved roles in gastrulation stage differentiation (epiblast activity) and germ cell specification (germ cell activity). Based on the expression patterns of different Oct4 homologues (POU5F1, 3) in African clawed frog (*Xenopus laevis*, Xl) and tammar wallaby (*Macropus eugenii*, Me), Oct4 homologues appear to have adapted lineage specific roles. Xlpou91 (pou5f3.1) and MePOU5F1 are expressed specifically in germ cells (germ cell specific POUV proteins), while Xlpou25 (pou5f3.2) and MePOU5F3 are expressed at high levels in gastrulation stage epiblast or ectoderm (epiblast specific POUV proteins). Here we ask if these expression patterns correlate with distinct functional activities in mammalian cell culture. We found that germ cell specific POUV proteins have the capacity to support self-renewal in Oct4-null murine ESCs, whereas epiblast-specific POUV proteins have only a limited ability to support self-renewal, and allow for differentiation to both trophoblast and primitive endoderm. While all Oct4 homologues appear to function in the reprogramming of fibroblasts to induced pluripotent stem cells (iPSCs), we observed marked differences in efficiency. We found that reprogramming efficiency could be augmented by either increasing POUV dose or by giving cells more time to reprogram me. The specific activity of POUV proteins in reprogramming appeared to correlate with their expression in the germ cell lineage. Taken together, our observations suggest that Oct4 has two distinct functions in development that have repeatedly become segregated in evolution: an epiblast function in supporting gastrulation stage progenitors and an independent function in germ cell specification. It is the germ cell specific activity of POUV proteins that best resembled the state present in ESCs and required to promote reprogramming to iPSCs.

P-219 The level of FGF signalling modifies shape and size of limb bones

Cela, Petra (Academy of Sciences of the Czech Republic, Brno, CZE); Krejci, Pavel (Masaryk University, Brno, CZE); Buchtova, Marcela (Academy of Sciences of the Czech Republic, Brno, CZE)

High variability in the arrangement of stylopod and zeugopod bones exists among species. In chicken and alligators, the ulna is much larger than the radius. This is in contrast to many other mammalian species, where the radius is the main load-bearing bone of the forelimb and the ulna can be significantly reduced in size or almost missing. Furthermore, overall shape of the humerus varies and numerous protuberances can be formed in some species. As fibroblast growth factors (FGFs) are key players in the processes of proliferation and differentiation during limb development, we experimentally manipulated FGF signalling in developing limbs to test its effect on bone modeling. We used loss-of-function and gain-of-function approaches, where FGF-receptor inhibitor PD173074 or recombinant FGF ligand (FGF1) were applied to the right chicken wing bud at stage HH20-22. Embryos were collected following 10-12 days of incubation and stained with Alizarin red/Alcian blue solution for skeletal analysis. FGF1 application caused shortening and thickening of the humerus. In some cases, there was a small protuberance on its side, which is ectopic to chicken, but resembles the tuberositas deltoidea of some mammalian species. The ulna was also shorter a thicker. The radius was bent as both epiphyses were in contact to the reduced ulna. The inhibitor treatment led to a shorter and thinner humerus as well as the partial or full absence of the radius, whereas the ulna remained without morphological changes. In conclusion, we found that alteration of FGF signalling affects the size and shape of long bones during limb development. Furthermore, we showed that a subtle modulation of FGF level leads to a reduction of one of the antebrachial bones. A similar modification of FGF signalling could play a role during limb evolution in vertebrates where different degrees of antebrachial bones reduction are seen.

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P-220 Towards an understanding of the genetic basis of phenotypic change

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Drosophila legs display variable patterns of trichomes (non-sensory cuticular actin protrusions). A region of the cuticle on the femur of the second leg termed the "naked valley" is free from trichomes. The size of the naked valley differs between species but is generally constant within species. *D. melanogaster* is an exception to this rule, as natural populations can exhibit different naked valley sizes. A genetic mapping approach was used to determine the loci responsible for this size variation. A region of approximately 25 kb that accounts for over 90% of the variation in the size of the naked valley was mapped to chromosome 3. One of the genes in this region encodes an intronic micro-RNA, mir-92a. mir-92a is capable of regulating in vitro and in vivo expression of shavenoid, a gene well known to be involved in trichome development. While the sequence of mir-92a is conserved in *D. melanogaster* strains with different naked valley size, its expression differs. This indicates that a regulatory element controlling mir-92a expression has evolved to give rise to different mir-92a levels. Differential expression of mir-92a thus leads to differential expression of shavenoid (and possibly other genes), which consequently determines the size of the naked valley. We have identified several putative enhancers of mir-92a and are currently testing which one might have evolved to lead to diversification of naked valley size.

P-221 The impact of gene loss on EvoDevo: Dismantling the retinoic acid genetic machinery in *Oikopleura dioica*

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The importance of the impact of gene loss in the evolution of developmental mechanisms is a crucial question in EvoDevo that remains unanswered. Until recently, gene losses had been often neglected because the proofs for a gene loss were negative and difficult to detect. The genomic era, however, offers the exciting opportunity to detect unambiguous cases of gene losses. The sequencing of the genome of the urochordate *Oikopleura* dioica has revealed an extraordinary genomic plasticity that led to extensive gene losses. *Oikopleura* is thus a suitable model to investigate the impact of gene losses in the evolution of the mechanisms of development. As a case study, we are analyzing the magnitude of gene losses that accompanied the dismantling of the retinoic acid (RA) genetic machinery (RAM) in *Oikopleura*. Our results exemplify the "EvoDevo inverse paradox", in which *Oikopleura* lost most RAM members, probably due to a reduction of developmental constraints that facilitated the dispensability of this morphogenetic system, without perturbing its typical chordate body plan. *Oikopleura,* however, also unexpectedly retained few retinoid metabolism genes that not only survived, but further expanded by extensive gene duplications. *Oikopleura*, therefore, appears as an attractive system free of classical RA-signaling for uncovering alternative functions of surviving members of the RAM, and to analyze the presence of novel retinoid compounds potentially important for embryonic development or tissue homeostasis in chordates.

P-222 New models of dipteran development: *Megaselia abdita* and *Clogmia albipunctata*

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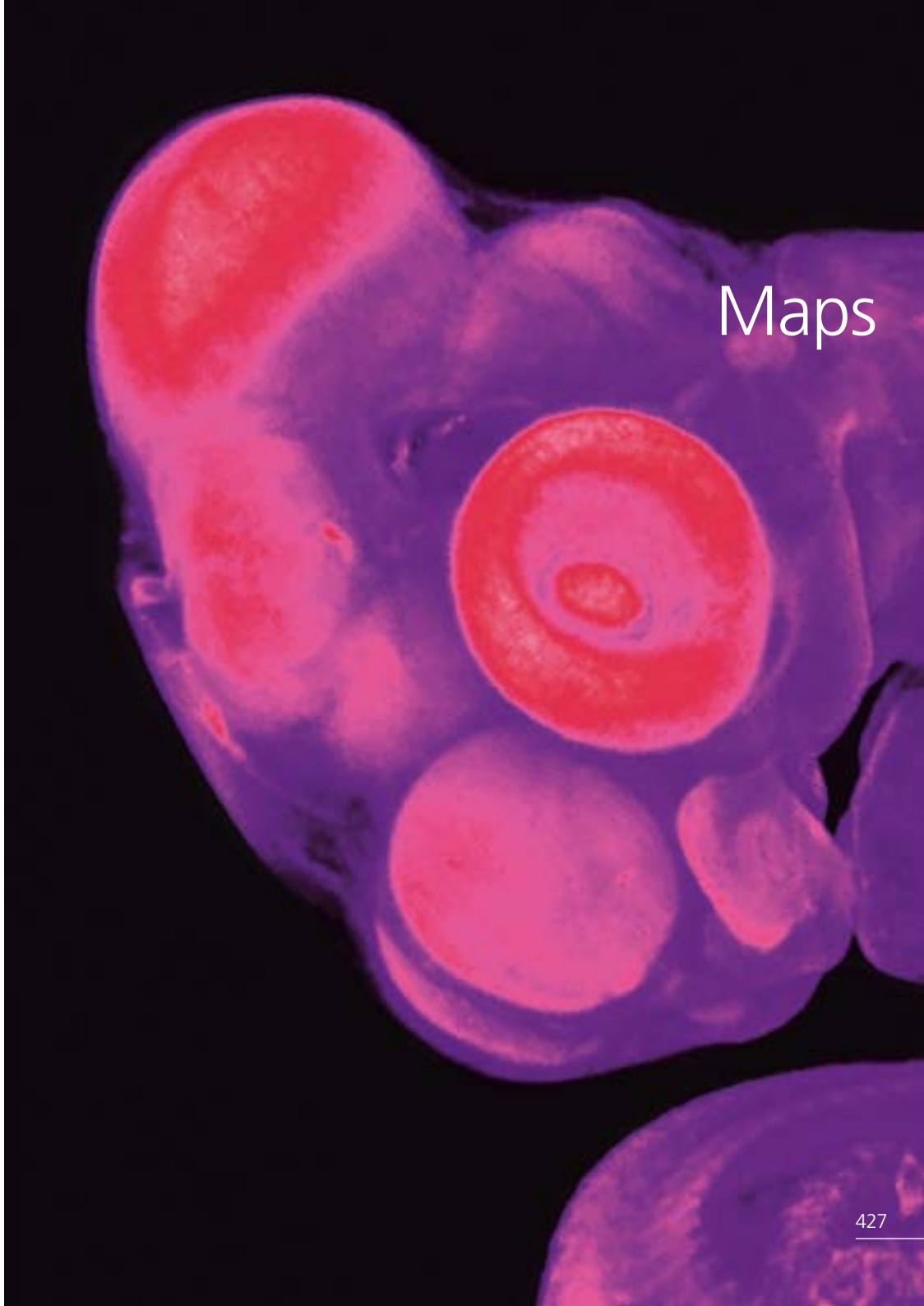
Model organisms such as *Drosophila melanogaster* allow us to address a wide range of biological questions. However, approaches using model systems need to be complemented by comparative studies for us to gain a deeper understanding of the functional properties and evolution of developmental processes. The establishment of new model organisms is crucial for this purpose. One of the first essential steps to establish a species as an experimental model is to carefully describe its life cycle and development. Here we provide a staging scheme and morphological characterisation of the life cycle for emerging non-drosophilid dipteran model systems: the scuttle fly *Megaselia abdita* and the moth midge *Clogmia albipunctata*.

Notes



Maps

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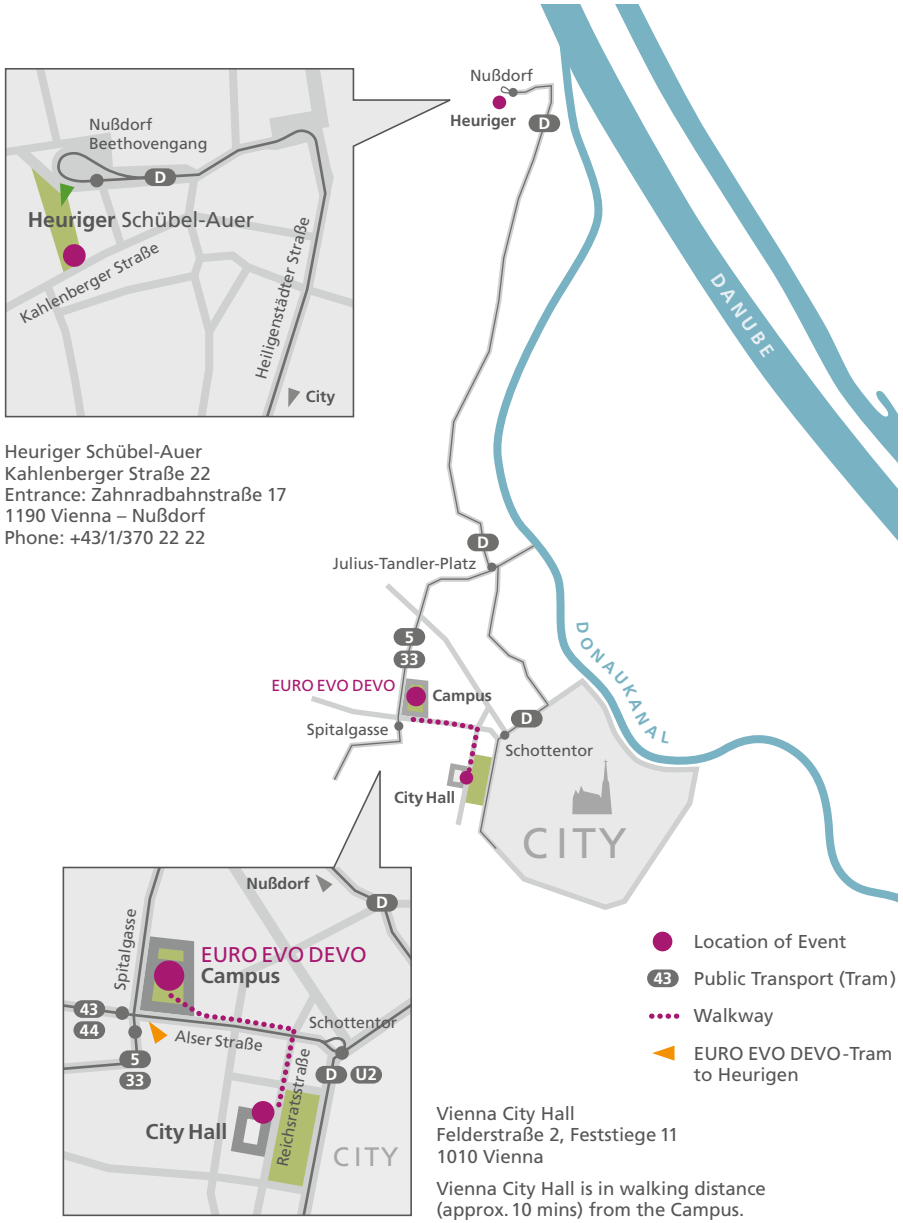

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