

segmentation clock of the zebrafish, a function not required in slower clocks such as the mouse.

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15-P019

Neurogenesis in the onychophoran *Euperipatoides kanangrensis* Joakim Eriksson, Angelika Stollewerk

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Our analyses of early neurogenesis in several representatives of chelicerates (e.g. spiders) and myriapods (e.g. millipedes) have revealed that the genetic network involved in recruitment and specification of neural precursors is conserved in all euarthropod groups. However, the expression pattern and function of these genes is adapted to the distinct morphology of neural precursor formation in each group. We observed several molecular and morphological characters in the developing central and peripheral nervous system of chelicerates and myriapods that cannot be found in equivalent form in insects and crustaceans. It is possible that these characters are shared derived characters (synapomorphies) of myriapods and chelicerates, providing the first morphological support for a clade uniting these two groups. However, they could also represent ancestral characters (symplesiomorphies) retained in myriapods and chelicerates and lost in the more derived insects and crustaceans.

We have therefore analysed neurogenesis in a representative of an outgroup to the euarthropods, the onychophoran *Euperipatoides kanangrensis*. We have identified Notch and Delta homologues in *Euperipatoides kanangrensis*. These genes are involved in specification of neural stem cells in insects. We show that they are expressed in a distinct pattern which is neither comparable to insects/crustaceans nor to chelicerates/myriapods. Furthermore, we have analysed the morphology of neural precursor formation by F-actin staining and light microscopic sections. The data suggest that at least some of the characters shared by chelicerates and myriapods are synapomorphies

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15-P020

Cell autonomous sexual development in birds

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Our understanding of sexual development in mammals is largely based on the paradigm established by the pioneering work of Alfred Jost and others in the mid-20th century. This established that, at a specific point in development, the sexually dimorphic

expression of a sex-chromosome gene induces the indifferent genital ridge to initiate gonadal development and that all other sexual development is a result of somatic cells responding to specific hormones. This principle is still widely accepted today and assumed to apply across vertebrates, although for many species the exact nature of the sex-determining mechanism and the identity of the sex-determining gene(s) have yet to be established.

In birds, sex-determination is thought to be dependent on either a dominant ovary-determining gene on the W-chromosome or a dosage mechanism based on the Z-chromosome. To help resolve this issue, we analysed three gynandromorph chickens that display a bilateral male:female asymmetry. For the first time, we demonstrate that gynandromorph birds are genuine male:female chimaeras, with tissues comprised of normal male and female diploid cells. This finding demonstrated that the sexual phenotype in birds is not dependent on gonadal products, but is largely cell autonomous and suggests that the mechanism of sex determination in birds is different from that seen in mammals. To investigate this possibility, we carried out a series of transcriptomic screens and experiments in which we generated embryos containing male:female chimeric gonads. Our analyses provide conclusive evidence that sexual development in birds does not follow the Jost principle.

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15-P021

Drosophila anterior determination is independent of Bicoid morphogenic functions but depends on activating versus repressing inputs on the level of target gene enhancers

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Bicoid (Bcd) is the anterior determinant of *Drosophila*. The mRNA of this homeodomain transcription factor is maternally deposited at the anterior pole, extends posteriorly and presents the basis for the anterior to posterior Bcd gradient in embryos. Bcd target genes are thought to be activated by specific threshold concentrations. Thereby, Bcd functions as a morphogen, instructing fields of cells to take on distinct fates. However, we have found that spatial boundaries of anterior target genes are also set in the absence of a Bcd gradient and depend on an antagonizing factor under control of the maternal terminal system. Its activated key component, the receptor tyrosine kinase Torso (Tor), down-regulates Capicua (Cic), a maternally provided transcriptional repressor, at the poles and thereby generates high Cic activity in the central region that decreases towards the poles. The spatial limitation of Bcd-dependent head gene expression is not dependent on the Bcd concentration gradient but on Cic activity which causes repression through specific binding sites in Bcd reactive enhancers, thus antagonizing activation by Bcd. While Cic is highly conserved, neither Bcd nor another anterior morphogen has been identified outside the Diptera. Thus, the emergence of the Bcd morphogen would require an extensive remodeling of anterior gene enhancers to become competent to react to distinct concentration thresholds

within the Bcd gradient. Our results allow us to conclude that Bcd does not function as a morphogen but rather is a newly evolved player in what is otherwise a conserved “anterior determinant system”.

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15-P022

Dynamics of cellular aggregation and differentiation during early development of transgenic crickets

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The cricket *Gryllus bimaculatus* is an emerging model organism for developmental and regeneration studies. In the cricket, the means for loss-of-function analyses of genes have been well established using RNA interference, but not for gain-of-function analyses. The *piggyBac* transposable element from the moth *Trichoplusia ni* encodes a DNA transposase that has recently been used to transform a number of insects, including the non-model insects. Thus, we have attempted to establish the method of genetic manipulation in the cricket using the *piggyBac* transposable element. We injected a plasmid containing a *piggyBac* element carrying *Gryllus* actin promoter-GFP marker and invitro synthesized mRNA encoding the *piggyBac* transposase into the cricket eggs. Transformed G1 crickets were obtained from 16% surviving individuals.

Using the GFP-expressing transgenic crickets, we succeeded to visualize cellular dynamics during blastoderm stages and early embryogenesis applying a live imaging technique. We found that in the cricket, the cellularization occurs during mid-blastoderm stage. In this stage, the cells are sparsely distributed in the egg and mitosis occurs asynchronously. After mid-blastoderm stage, the cells of posterior half of the egg synchronously migrate to the posterior region to form a germ anlage consisting of bilateral aggregates, while remaining cells do not migrate. Our data suggest that the embryonic primordium is formed dynamically by cellular aggregation and differentiation. We are conducting RNAi analyses using transgenic crickets to elucidate the mechanisms of early development. The results will be reported.

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15-P023

The evolution of neural induction in deuterostomes

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Knowledge of the development of the brain continues to grow, yet the evolutionary origin of the central nervous system (CNS) remains unknown. Organisms as diverse as flies and mammals have a centralized nervous system, and share many neural inducing and patterning genes, yet they are also markedly different from one another. These differences raise the question of whether deuterostomes and protostomes share a common ancestor or if they emerged independently of each other. One way to address this is to study development of the nervous system of the hemichordate *Saccoglossus kowalevskii* which has a basic chordate structure but the developing embryos have a basi-epithelial nerve net instead of a centralized nervous system. Hemichordates are a sister group of the chordates and recent work on axial patterning in the hemichordate demonstrate similarities with vertebrates indicating that *Saccoglossus* is critical to our understanding of the evolution of neural induction and patterning. To determine what drives the induction of neural cells, we analyzed the expression pattern of common markers of neural, neuronal and epidermal cells. In this way we can determine when the neural cells are induced, when differentiation is initiated and the pattern of the nervous system as development proceeds. Using chemical inhibitors of FGF and Notch-Delta signaling, we address the role of these signal pathways on neural induction and differentiation. Identification of the molecular events involved in the induction and maintenance of the hemichordate nervous system allows us to better understand the evolutionary history of the centralized nervous system.

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15-P024

Retinal function in the closed eyes of the Iberian mole

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The evolution of the eye has fascinated biologists for more than a century. Reduced visual systems are common in animals adapted to live in the dark. Fossorial mammals have a wide variety of dysgenic eyes. We have studied the development of the eyes in the Iberian mole *Talpa occidentalis*. It was found that these animals have small but surprisingly well developed eyes that remain permanently closed under the skin even in the adulthood. There are no eyelids. A laminated retina is formed, in which all normal cell types are present. However the retina seems dysgenic. There is a disruption of the inner plexiform layer, in which numerous ectopic ganglion cell bodies are randomly distributed. Most of these cells express PAX6 and many of them contain high amounts of melanopsin. Rod- and two cone-opsins, shortwave-sensitive (S) and middle-to-long-wave-sensitive (M), are present in the photoreceptor layer, but there are also some opsin-expressing cells displaced to other layers. The presence of these opsins, together with the fact that Dil labelling shows that ganglion cell axons project contralaterally to the visual cortex of the brain, indicates that these animals are capable of detecting and processing light stimuli. Moreover, the high proportion of retinal ganglion cells expressing melanopsin leads us to propose