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Ancient amber meets modern methods: Using micro-CT scanning to describe new species of *Spalangiopelta* from Baltic amber

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Spalangiopelta (Hymenoptera: Pteromalidae) is a small genus of parasitoid wasps that has received little attention despite the widespread distribution of its extant species. The fossil record of the genus is restricted to a single species from Miocene Dominican amber. We described two new species of *Spalangiopelta* from Baltic amber, which significantly extend the minimum age of *Spalangiopelta* to the Upper Eocene. Phylogenetic relevance and functional morphology of two wing characters, admarginal setae and the hyaline break, are discussed.

3D models assisted in the descriptions, the determination of character states and the construction of an identification key for the females of all currently known *Spalangiopelta* species. The models were reconstructed from X-ray μ CT scans, pre-segmented manually in Amira and completed by semi-automatic segmentation in Biomedisa. This method has proved to be an invaluable addition to classic techniques in the description of amber fossils as the pivotable model eliminates physical barriers such as refraction or reflections. It further allows for the digital removal of inclusions such as air bubbles or plant particles that conceal relevant structures of the specimen.

Structural and functional morphology of the spinning apparatus of cribellate spiders with special focus on their capture thread production

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The properties and structure of spider threads are highly diverse depending on their function. To capture prey, cribellate spiders produce threads of various combined fibre types that rely on adhesion forces such as van der Waals and hygroscopic forces in combination with capillary forces. These forces are mainly caused by the incorporation of nanofibres. Other fibre types perform specifically mechanical tasks. The spinning apparatus is important regarding the production of these complex threads. Specialised and highly manoeuvrable, paired structures on the posterior end of the opisthosoma called spinnerets allow the interaction of different fibre types which exit the body through spigots located on the spinnerets. To produce nanofibres, cribellate spiders additionally own a specialised structure beside spinnerets, the cribellum, and a comb-like structure on their hind legs to pull them out. However, little is known about movement interactions of the spinning apparatus connecting different fibres into one thread. Therefore, we examined the morphological features of the spinning apparatus with special focus on the movement sequences in different cribellate spiders (*Amaurobius similis*, *Deinopis subrufa*, *Eresus walckenaeri*) during cribellate capture thread production. Our results reveal differences in spinneret size, structure and movement velocities between the examined species.

The prothoracic defensive glands of stick and leaf insects

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Phasmatodea (stick and leaf insects) are well known for their ability to disguise themselves as twigs, leaves or bark. Despite this and many other beneficial defensive strategies, many phasmatodean species can actively spray defensive chemicals upon disturbance from predators or parasites by the use of prothoracic defensive glands. Size and structure of these glands differ greatly in different phasmatodean species. Additionally, there is a wide range of chemical components in the defensive secretion. The advantage of the secretion ranges from irritation of the mucous membranes and unpleasant odor to little or no perceptible effect, which probably is attacker/parasite-dependent. To this day, only twelve species have been studied regarding the morphology and/or chemical content of the defensive glands. Our main goal is to generate a profound information base for the anatomical and biochemical diversity of the prothoracic defensive glands across a broad taxon sampling of Phasmatodea in a phylogenetic framework. We use μ CT Scans to reconstruct the anatomy of the defensive glands and gas chromatography/mass spectrometry to detect the chemical substances of the defensive secretion.

Morphology and closing mechanism of the mandibular gland orifice in ants (Hymenoptera: Formicidae)

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The mandibular gland of ants releases chemical compounds with various functions. The morphology of the gland is well investigated in several species, but the mechanism of secretion release was not explicitly addressed so far. To clarify this question, we examined the gland orifice anatomy in species of 14 ant subfamilies employing different techniques. The orifice is located on an area called mandalus with variable external shape. The internal organization is remarkably congruent across all investigated species. The mandalar cuticle is always connected to the mandibular gland duct by a lamella, visible as characteristic anchor-shaped structure in cross section. No muscles were found in association with the orifice, nor with any other part of the mandibular gland. However, the mandalar base is connected to the prepharyngeal sucking pump by a cuticular ligament. Additionally, it is continuous with the conjunctiva connecting the mandible to the head capsule. We propose that retraction of the sucking pump by the muscle *M. tentorio-buccalis*, potentially in concert with opening of the mandible, stretches out the ligament and thus pulls on the mandalus and mandalar lamella to open the gland orifice and allow for secretion release. This hypothesis is congruent with findings in other aculeate Hymenoptera.

Compensatory suction feeding in dragonfly larvae? Evidence from computational fluid dynamic simulations

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Dragonflies and damselflies (Insecta: Odonata) capture prey with a fast moving, highly modified mouthpart, the prehensile labial mask. Within dragonflies (Odonata: Anisoptera) two basic shapes of the prehensile labial mask have evolved with an either flat and slender or concave - spoon-shaped - distal segment (prementum). While the former is a pure grasping device, the latter is also capable of scooping up smaller prey items to retain them inside the cavity. Although some hypotheses exist about the topic, the hydrodynamics of the prehensile labial mask have not been investigated so far. Based on 3D μ CT reconstructions, we used computational fluid dynamic (CFD) simulations of the prementum to understand the relationship between the shape of the prehensile labial mask and its hydrodynamic properties. Our results suggest that both types are highly streamlined and generate a low-pressure area, likely leading to an effect analogous to compensatory suction feeding. This is a well-studied strategy in different vertebrates, to mitigate the formation of a bow wave which could alarm or even displace prey in front of the predator. Studying the hydrodynamics of fast prey capturing movements might be an interesting concept for technical application in small scale technical underwater grasping devices.

Morphological variation in the larval malpighian tubules impetus social adaptations in ants

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The excretory system in insects and terrestrial arthropods consists of structure called malpighian tubules. They are analogous to vertebrate renal tubules or nephrones. The role of malpighian tubules is maintaining constant internal environment through the elimination or segregation of unnecessary substances present in the hemolymph, and the retention or reabsorption of useful substances to the body. Social insects are highly evolved insect groups with clear reproductive division of labour. The increased complexity at organism level by a major evolutionary transition from solitary to social life increased their survival which eventually led to their ecological success. Here, we studied the morphological variation of the malpighian tubules across the worker and queen larvae of a eusocial ant, *Oecophylla smaragdina* (Asian weaver ant). The malpighian tubules in the workers are comparatively thicker and convoluted. Whereas that of queen larvae are relatively long, smooth tubules that end blindly in the abdominal cavity and are highly branched. The variation in the Malpighian tubules among the castes indicates task based social life adaptation in the ant colony.

Giant heads suggest ecological convergence in platygastrine parasitoid wasps

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Platygastrinae (Hymenoptera: Platygastridae) are parasitoids of gall midges (Diptera: Cecidomyiidae). The biology and ecology of most species are unknown, and most genera remain in a state of taxonomic disarray. In the genus *Synopeas* Förster, a preponderance of vague species descriptions based on subjectively evaluated characters constitutes a severe impediment to identification. Although many species are difficult to distinguish morphologically, a few species exhibit bizarre and extreme morphology. We recently discovered a new species of *Synopeas* from New Guinea which has an unusually large head. The new species description elevates the standard of platygastrine taxonomy, including COI barcode data, gall host plant data, and machine-readable morphological characters. Using microphotographic techniques and scanning electron microscopy, we examined the head shape and compared it with the heads of other platygastrine species. A similar head is found in species of *Inostemma* Haliday which share some ecological associations, as well as in species of *Platygaster* Latreille whose ecology is unknown. The functional implications of this unusual head shape are explored. Further lines of inquiry are proposed.

Morphology and systematics of the boring bryozoan family Penetrantiidae

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An endolithic lifestyle within mineralized substrates has many advantages and evolved multiple times in various phyla including Bryozoa. The family Penetrantiidae includes one genus with approximately a dozen species and colonizes predominantly shells of living and dead molluscs. Based on the presence of an operculum and other morphological characters, penetrantiids were debated to be either ctenostome or cheilostome bryozoans. Consequently, the aim of this study is to analyze the morphology of the genus *Penetrantia* with modern methods such as confocal laser scanning microscopy and 3D reconstruction based on serial sections. So far *Penetrantia parva* (New Zealand), *Penetrantia concharum* (Sweden) and a potential new species (French Atlantic) were analyzed. First data on the musculature associated with the operculum and the digestive tract show strong similarities to other ctenostome bryozoans, which is also supported by first preliminary results of a phylogenetic analysis based on COI and 18S sequence data. Increased taxon sampling and a broader multigene phylogenetic analysis will shed even more light on certain morphological traits and consequently on the systematics of this little investigated bryozoan family.

Comprehensive single-cell atlas of a bivalve larva and developmental decisions of molluscan cell lineages

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Mollusks (bivalves, gastropods, cephalopods, polyplacophorans and their kin) are known for their morphological innovations and lineage-specific plasticity of homologous characters (e.g., radula, shell, foot, neuromuscular systems), raising questions concerning the cell types and the molecular toolkit that underlie this variation. Here, we report on the gene expression profile of the trochophore larva of the invasive freshwater bivalve *Dreissena rostriformis*. We generated transcriptomes of 682 individual cells and identified nine transcriptionally distinct cell populations. To annotate these cell populations, we examined the gene sets that characterize each individual cluster and compare these to gene expression data previously reported from other lophotrochozoans. Genes expected to be specific to certain tissues, such as *Hox1* (in the shell field), *Caveolin* (in prototrochal cells), or *FoxJ* (in cilia-bearing cells). This corroborate that the recovered cell clusters reconstruct expression profiles for different tissues previously described in morphological studies. Furthermore, developmental trajectory analyses group together clusters that share an ectodermal origin, resembling the cell fate map from embryonic stages. Altogether, this dataset provides a molecular atlas of gene expression underlying bivalve-specific tissues, whilst establishing a foundation for further comparative studies and adding another layer of knowledge regarding the plasticity of molluscan body plans.

New unusual features of proboscis in females of *Bonellia viridis* (Annelida: Bonelliidae)

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Echiurida is a small group of marine annelid lost their segmentation. They lead a hidden lifestyle and are extremely difficult to access for morphological studies, especially for investigation of ultrastructure. Despite the fact that *Bonellia viridis* is the main object for morphological research in the echiurids, many details of the anatomical structure still remain unclear. New data on the anatomy and ultrastructure of *B. viridis* will help to find the main ways of evolution of the organ systems within Echiura. Epithelium of proboscis lacks the basal lamina and therefore can not be regarded as true epithelium. The same concerns the coelothelium, which lines the coelomic canals. This is probably caused because of bonellin producing cells, which are able to penetrate into epithelium and into coelomic lumen. Cells of dorsal side of proboscis are able to absorb small particles from the environment. The nerve tracts innervating large epidermal glands of the proboscis are described for the first time. According to our data, the wall of the axial blood vessel is formed by myoepithelial cells, which form numerous thin projections directing into the lumen of vessel and into the connective tissue. The study is supported by Russian Foundation for Basic Research (#20-04-00096).

Liquid uptake in Vespinae (Vespidae, Hymenoptera) – The morphology and function of mouthparts, hypopharynx and pharynx

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The labiomaxillary complex of Vespinae evolved to efficiently forage various solid and liquid foods, such as animal proteins, carbohydrate rich fluids and water as well as woody fibres for nest construction. Before liquids are swallowed and transported into the crop, particles are filtered out. This study examines the morphology and function of the mouthparts and alimentary tract, focussing on this filtration process. The feeding organs, preoral cavity and pharynx, are studied using μ -CT, 3D reconstructions of semithin sections, SEM and feeding experiments with glass beads in workers of *Vespula germanica*. To test for filtration, barium sulfate has been used as a fluid contrast agent, a method rarely used in entomology. The results indicate that particles bigger than 212 μm are filtered by the mouthparts. The pharynx bears interlinked microtrichia arranged in rows oriented towards the infrabuccal pocket. Together with the cuticle structure of the epipharynx and the hypopharynx, this enables an additional filtration of particles bigger than $\sim 100 \mu\text{m}$. This could be the maximum size of particles, which are able to pass the narrow wasp waist. For the first time a detailed illustration of the multi-layered filtration system formed by the mouthparts and the preoral cavity and its function is presented.

Comparative morphology of the venom delivery system of selected atractaspid, elapid and viperid snakes using micro-CT based 3D modulation

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Despite a century and a half of morphological and phylogenetical research, the evolution of the venom delivery system in snakes remains unresolved. While venom glands and associated teeth share the same developmental origin, their structure is fundamentally different on an interfamilial level, making it difficult to trace their evolution, especially with the remaining lack of a strongly supported phylogenetic tree. The present work dealt with the morphology of the venom delivery system in the snake families Atractaspididae, Elapidae and Viperidae. To this end, *Atractaspis irregularis*, *Elapsoidea nigra* and *Atheris choleris*, which were chosen as respective representatives for these families, were stained with phosphotungstic acid and scanned using micro computed tomography. The images were then processed into 3D models, which were used to describe and compare the morphology and inner structure of the venom glands. The results generally confirmed those of previous studies that relied on other methods (dissection, histology), demonstrating that MicroCT based 3D models are a reliable non-destructive method for depicting the form, structure and natural position of the venom delivery system. Additionally, the possible evolution of the venom apparatus was discussed. Based on all available morphological data, including the results of this work, the newest phylogenetic tree and the latest divergence time estimates, it was inferred that the venom delivery system of Endoglyptodonta might have evolved convergently on basis of a basal member of the Caenophidia, which possessed a primitive form of a venom gland.

The morphology of lophopodid Phylactolaemata: Implications for the neuromuscular groundpattern

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Bryozoans are colonial, sessile, suspension-feeding lophotrochozoans. The solely limnetic Phylactolaemata are considered as sister taxon to the vast majority of predominantly marine Myolaemata. The former includes only 70-80 species subdivided into 6-7 families but are crucial for groundpattern reconstruction. Especially the morphology of the early branching Lophopodidae (*Asajirella*, *Lophopodella*, *Lophopus*) is little investigated. Since there is almost no data on *Asajirella*, the aim of this study is to analyse the neuro-muscular system of *Asajirella* and lophopodids in general, using immunocytochemistry, confocal microscopy, and 3D-reconstruction. The myoanatomy shows tentacle muscles interconnected to the lophophore arm muscles, as typical for taxa with large lophophores. Duplicature bands connect the body wall with the tentacle sheath and not the diaphragm as previously claimed, which supports this character being present in all phylactolaemates. A cerebral ganglion with a large lumen is present in all investigated lophopodids. In addition, conical horns ascend into the phylactolaemate-specific epistome. Ultimately, the epistome of all investigated genera is a short bulge above the mouth opening. In summary, the new data support the characters found in the phylactolaemate groundpattern, but also allow to characterise Lophopodidae by novel, apomorphic traits.

Fingerprint-like nanostructure on a spider's hind leg prevents sticking to nanofibers

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Nanofibers attract attention due to their advantageous surface-to-volume ratio and the variety of possible applications. However, their production remains challenging, as such fibers adhere to almost all surfaces due to van der Waals forces. However, engineers can learn from nature's specialists in fibre processing: cribellate spiders. These spiders weave thousands of nanometer-thick fibers into their threads to catch and hold their prey. They use a comb-like structure on the hindmost metatarsi, called calamistrum. We were able to demonstrate that the calamistrum is essential for the spiders to handle nanofibers without sticking to them. This anti-adhesive property is due to a fingerprint-like nanostructuring on the setae of the calamistrum, which effectively reduces van der Waals forces by preventing fibres from smoothly adapting to its surface, thus minimizing the contact area. The successful biomimetic transfer to artificial surfaces highlights the functionality of the structure and presents a first technical application of this innovative and so far, unique structure.

An uncommon defense mechanism - A characterization of the glue in four different centipede species

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Centipedes are known for their hunting behavior with venomous forcipules. Beside this predation strategy, some centipedes defend themselves through a fast-hardening glue produced by epidermal glands, so-called sternal glands, located on the ventral surface of each trunk segment. However, little is known on the chemical composition of the glue and interspecific morphological differences of the glandular. Thus, we investigated four different centipede species (*Henia vesuviana*, *Henia illyrica*, *Haplophilus subterraneus* and *Strigamia maritima*), and preliminary results reveal a clear difference in size and distribution of these glands. By energy dispersive X-ray spectroscopy, a higher sulfur concentration was measured in the glue of *S. maritima*. Histochemically, the secretions of three species contain acidic mucosubstances, glycoproteins and basic proteins. Furthermore, lectin affinity tests on the native glue showed a clear specificity for a few sugar moieties (mannose, N-acetyl-D-glucosamine and lactose/ β -galactose). Chemical evaluation by polyacrylamide gel electrophoresis showed unique protein band patterns for each of the four species. Although the sternal glands share common characteristics, we found variances in their morphology and also chemical properties. Different abiotic factors (habitat, predators and food source) may be considered as one potential reason for species-specific features of the centipede glue.

The first 3D reconstruction of the coelomic system in *Novocrania anomala* (Brachiopoda, Craniiformea)

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The organisation of the coelomic system in bilaterians is important in the light of such a fundamental problem as body compartmentalisation of the last common bilaterian ancestor. First complete 3D reconstructions of the coelomic system of *Novocrania anomala* has been done using serial sectioning. Coelomic system of *N. anomala* consists of five main parts: two large brachial canals, tentacle base coelom (i.e., perioesophageal coelom and paired small brachial canals), paired frontal chambers, main trunk coelom, and paired posterior adductor chambers. Thus, accordingly to our data, *N. anomala* possess “quinquepartite” coelomic system, which has never been described in brachiopods before. Relations between several coelomic cavities are still unobvious, especially in case of the frontal chambers and posterior adductor chambers. Anatomy of the coelomic system has a lot in common in craniiformes and rhynchonelliformes. The anal chamber, which was described by F. Blochmann in *N. anomala* in 1886, has not been discovered by the method of 3D reconstructions. Some peculiarities of the coelomic system organisation in *N. anomala* may be considered as a support for “brachiopod fold” hypothesis. This study is supported by Russian Science Foundation (#18-14-00082).

Transitions in allometry reveal the evolution of worker caste polymorphism in farming fungus Ants

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Worker polymorphism in ants is defined by intraspecific variation in morphology producing task specialization. In 1953 Edward Wilson described the diversity of worker subcastes by the degree of dissociation of the head regarding thorax length through allometry, thus, the level of correlation between these two traits is explained by the level of deviation from linearity. In this study, I am addressing the evolution of worker caste in ants as changes in their head to body allometry. Hence, I focused on species of farming fungus ants, this clade represents an evolutionary transition from the life of hunters to farmers subsisting on cultivated fungi. Firstly, we have sampled individuals from several species across the group. For all the species the natural log of thorax length (TL) was plotted against the natural log of head width (HW), a model linear regression was tested for all species, and for those that did not follow a strict linear regression, we tested several models to explain better the allometry pattern. The analysis has shown a phylogenetic transition of head-to-body allometry in these species. Specifically, across the phylogeny there is an increase of size variation and deviation from linearity that ultimately could explain the evolution of polymorphism in the group.

References.

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The gland system of *Latia neritoides* (Gastropoda) in regard of its unique defence mechanism

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The New Zealand endemic freshwater gastropod *Latia neritoides* has a unique defence system: it releases an adhesive, luminescent mucus when threatened by predators. Some information is available on this bioluminescence while its adhesive properties are still unexplored. To understand the mucus composition, chemical analyses of the mucus were performed. In addition, all secretory glands in the sub-epithelial foot layer were characterized on morphological and histochemical level to correlate the mucus components to the different gland types. By this the study aims to determine relevant glands, involved in the production of this unique defence system.

The results reveal two gland types in the lateral area, while a third type is located in the ventral foot layer. The three gland types differ histochemically, while lectin affinities are similar among all of them. Element analyses indicate that the two lateral glands contain different amounts of sulphur, magnesium and phosphorus.

At present it is not yet clear, whether lateral glands contribute to the production of the defence mucus. Transcriptomic data are in progress to identify and further characterize the proteins of the defensive mucus on a molecular basis.

Long live the king? Skull morphology and intrasexual conflict in the eusocial Ansell's mole-rat

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African common mole-rats of the genus *Fukomys* combine a monogamous mating system and pronounced sexual size dimorphism; a pattern highly untypical for mammals. At the same time, they live in cooperatively breeding groups composed of reproductive and non-reproductive members of both sexes. Here we explore patterns of morphological differentiation in skulls of Ansell's mole-rats (*Fukomys ansellii*) by means of multivariate analysis of linear skull measurements combined with a 2D shape analysis of cranium and mandible. Compared to females, males display larger skulls relative to body size and show an expansion of the facial portion of the cranium, while reproductive status was not found to have an effect on any of the traits studied. We also show that species of *Fukomys* obey Rensch's rule of scaling in sexual body size dimorphism, which is deemed indicative of intense male intrasexual competition. Our results point to a greater role of male-male conflicts in *Fukomys* than is traditionally assumed but which is also supported by preliminary genetic field data.

Lophophore innervation in *Flustrellidra hispida* (Bryozoa)

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Ctenostome bryozoans are unusual gymnolaemates and their anatomy probably reflects ancestral state of bryozoans in general. Our research is focused on the peculiarity of the lophophore innervation in *Flustrellidra hispida*. Immunocytochemistry and confocal laser scanning microscopy revealed the presence of outer nerve ring, which gives rise to the tentacle nerves that are associated with the groups of outer perikarya. The outer nerve ring has been formerly described in some ctenostomates and cyclostomates, but is not connected with tentacle nerves. Formerly, the outer nerve ring was described in some ctenostomates and cyclostomates, but it has no connection to any nerves. The discovered feature of lophophore innervation of *F. hispida* allows to suggest the evolutionary transformation from a phoronida-like ancestor lophophore, bearing prominent outer nerve ring with numerous tentacle nerves emanating from it, to the complicated bell-shaped lophophore of *F. hispida* with well pronounced outer nervous ring bearing a few tentacle nerves. Next one in this hypothetical row will be the lophophore of the other ctenostomates and some cyclostomates with an outer nerve ring having no connection to any nerves, and cheilostomates lophophore, lacking an outer nerve ring at all. The study is supported by Russian Science Foundation (#18-14-00082).

Functional morphology of the molluscan radula – current approaches and future perspectives

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One molluscan autapomorphy is the radula, a thin chitinous membrane spiked with teeth, mechanically processing and gathering all varieties of food types. Since the radula is an interface between the organism and food understanding its functional principles may contribute to the understanding of molluscan ecology. Usually in literature, pure radular tooth morphology is examined whereas, for a deeper insight into functionality, the complex interplay between teeth, their underlying supportive structures, and their interaction with the target surfaces must be studied.

We here present an overview of our series of experiments revealing (1) gradients of mechanical properties in individual teeth and (2) role of water content in the mechanical behaviour of teeth. Then we discuss how these material properties contribute to a uniform stress distribution along individual teeth and between teeth while foraging. Furthermore, stress and strain simulations, using FEA incorporating experimentally obtained mechanical properties and 3D-morphology, are introduced. Finally, original experimental approach, determining the radula-substrate contact areas, is presented, and the underlying mechanisms preventing structural failure in radulae are discussed.

Our combination of approaches represents a further step towards the transition from theoretical, descriptive morphology to the physical world. As future perspective, we briefly introduce our ideas of building 3D printed realistic models of radulae, designed on the basis of our morphology data with the possibility to directly test hypotheses about functional principles in radulae.

Comparative analysis of mandible morphology: The curious case of *Oecophylla smaragdina* and *Cataglyphis longipedem*

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Mandibles can be considered to be one of the most vital tools for the survival and success of ants, having extensively evolved into diverse forms best suited for their modes of life. Mandible construction have been reported to be heavily dependent on the respective ecological niche and habit of the ant. Here, we test this hypothesis and try to understand the mechanisms involved in their diversification. In this study, we have compared the external morphology and zinc content of mandibles of two ants with different life modes and diets, *Oecophylla smaragdina* and *Cataglyphis longipedem*, using scanning electron microscopy and energy dispersive X-ray spectroscopy. We show that ant mandible size, shape and number of teeth vary significantly in co-relation to their individual habits, i.e. *O smaragdina*'s actively predatory group foraging and *C. longipedem*'s solitary scavenging. Preliminary results also portray that despite differences in external morphology, the zinc content appear to be the same. Thus, this study might prove to be instrumental in evaluating the various physical mechanisms involved in the evolution of insect mandibles for their defined function.

Morphology of the first discovered deep-sea phoronid

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Phoronids are benthic marine invertebrates from the Lophophorata clade. They have a worldwide distribution and can form dense benthic aggregations, where they act as ecosystem engineers. Paraffin histology, serial sectioning, scanning electron microscopy, and 3D reconstructions are used to describe and identify phoronids, which have been collected in the Sea of Okhotsk from up to 1551 m depth. Identified species belongs to *Phoronopsis* genera based on the presence of epidermal fold (a collar) at the base of the lophophore. The lophophore is transitional to spiral, number of tentacles is 120-150. Longitudinal muscles are feather-like; there is a single left giant nerve fiber. Species is dioecious, oocytes are mature in May, lophophoral organs are small and membranous. Metanephridia have ascending and descending branches of canal and one funnel, that opens in both oral and anal coelomic chambers. Morphological description of deep-sea phoronids cannot be fully attributed to known phoronid species. As before phoronids were found only on depths not exceeding 400 meters, this discovery is the first phoronid living on depth up to 1.5 km and the first record of phoronids in the basin of Sea of Okhotsk. Our research aids in the effort of documenting phoronid diversity, distribution, and ecology. The study is supported by Russian Science Foundation (#18-14-00082).

The prerequisites of 'pelvic brooding': A specialized reproductive strategy in Sulawesi ricefishes (Beloniformes; Adrianichthyidae)

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Reproductive strategies are complex traits, as they often combine morphological, physiological and behavioural adaptations. Being able to observe such multi-layered aspects of a species' life-history coming together in a specific set of traits, offers the unique opportunity to investigate this evolutionary interplay. In ricefishes (Beloniformes; Adrianichthyidae), a group of small freshwater fishes found in East-Asia, the vast majority of species are known to be transfer brooders. In these fishes, females deposit a cluster of eggs on submerged substrates shortly after spawning. However, for two ricefish lineages endemic to the island Sulawesi, a distinct reproductive strategy called 'pelvic brooding' is described. Females of pelvic brooding species (*Adrianichthys oophorus*, *Oryzias eversi*, *O. sarasinorum*) carry the fertilized egg-cluster for about two weeks until fully developed offspring hatch.

In a comparative approach involving species of both reproductive strategies, we employ high-resolution μ CT-imaging, morphometrics and histology to identify the morphological prerequisites associated with this unique reproductive strategy. We found similar adaptations in the two lineages of pelvic brooding ricefishes and are interested in the so-called 'plug structure', modifications of the ribs and conspicuously elongated pelvic fins in female pelvic brooders.

Capture of inducible defences and their costs in the model organism *Daphnia*

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Daphniids are textbook examples of phenotypic plasticity and especially for inducible morphological defence for decades now. Nonetheless, the three-dimensional morphology, the exact functional mechanism as well as the costs at which the increased survival comes, remained largely unknown. We are now able to leap forward using a combined morphometric, behavioural and digital simulation assay. For this purpose, we used a three-dimensional scanning technique based on confocal laser scanning microscopy to detect all morphological features the model species *Daphnia longicephala* expresses in presence or absence of the predatory heteropteran *Notonecta*. With this data we created an average 3D-model of predator-exposed and naive *D. longicephala* each, which allowed measuring increase or decrease of body surface and volume. We also employed accurate 3D-Video Tracking to determine realistic swimming velocities of both morphs. Finally, we merged these findings in virtual simulations of the daphnids movements, which enabled us to determine, for example, drag forces and energy consumption. For the first time, this approach allows us to measure the trade-offs between the defensive morphology and respective costs for locomotion, which can be calculated also for other species. This will help to expand our knowledge concerning mechanism and costs of these exciting inducible morphological defences.

Using micro-computed tomography to study the skeletal anatomy of adult *Aleiodes arnoldii* Tobias, 1976 (Insecta: Hymenoptera, Braconidae)

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The Braconidae is one of the biggest families of hymenopterans with over 21,000 described species worldwide. *Aleiodes Arnoldii*, as one of the parasitoid wasps of the family Braconidae, parasitizes moth's larvae; and have an important role in controlling the population of moths in agricultural and forest ecosystems. Here we describe an anatomical study of the external morphology using micro-computed tomography (Micro-CT) with 3D visualization of anatomical features. This technology enabled us to show internal anatomical features in detail. The samples were scanned at the Topo-Tomo beamline in KIT light source, Germany. This is the first complete 3D reconstruction of the anatomy of a parasitoid wasp and includes a 3D anatomical atlas that can be explored with android devices. This result improves the basis for most taxonomic work on the braconid's group. Volume Rendered images and videos represent a significant advance in the knowledge of parasitoid wasp anatomy. The 3D anatomical atlas constitutes a unique and beneficial tool for future research and as educational aids.

Functional relationships between beak shape and trophic niche in sympatric cephalopods

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The morphology of species is directly linked with their role within ecosystems. In this context, variation in the shape of anatomical structures used for prey capture and feeding can be related to foraging functions and thus may explain the existence of differences in the trophic niche between species. Most cephalopods are carnivorous and active predators. Their beaks are one of the few hard structures from their anatomy and can be suspected to be affected by selective biological functions as predation and food processing. Therefore, a direct link between beak shape and the kind of prey consumed by different cephalopod taxa is hypothesized. In this study, we investigated the functional relationships between beak shape and trophic niche occupied by 20 cephalopod species, including cuttlefish, squid and octopus coexisting in the Mediterranean Sea. Here we combined geometric morphometrics to accurately characterize beak shape and stable isotopes to obtain a detailed profile of trophic niche. The results revealed that beaks differed noticeably in their shape across taxa, varying extensively among functional groups. Similarly, the isotopic niche also differed between some of the species, suggesting the presence of trophic partition. The combination of both shape and trophic position apparently indicates a functional relationship between the beak shape and the trophic niche of some of these species.

Uncovering the morphological changes in the reproductive system of the female pelvic brooder *Oryzias eversi*

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A unique reproductive strategy called pelvic brooding is known for three ricefish species (Adrianichthyidae) endemic to freshwaters in Sulawesi. In pelvic brooding species, a bundle of eggs stays attached to the female, via long filaments that emerge from the genital opening, until the juveniles hatch. In contrast, in transfer brooding ricefish species, the eggs are deposited a few hours after spawning. In the present study, we investigated morphological adaptations in the reproductive system of female pelvic brooding *Oryzias eversi*. We sampled females at several time points over the reproductive cycle and generated histological sections. Over the course of egg carrying, a tangled mass of attaching filaments, epithelia cells, blood capillaries and collagen tissue becomes a compact plug-like structure in the anterior part of the oviduct. Moreover, oocyte maturation in the ovary seems to be suppressed and the structure of the ovarian wall changes during this time. Our results demonstrate a striking resemblance of the adaptations to pelvic brooding in the reproductive system of *O. eversi* with the pelvic brooding species *Oryzias sarasinorum*.

Ilia variability of *Plateosaurus* specimens from Frick (Switzerland) - preliminary results.

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Plateosaurus is a basal sauropodomorph and the most abundant late Triassic dinosaur in Europe. Therefore, it is not surprising that *Plateosaurus* was the subject of several investigations and publications dealing with its ontogeny, morphology, phylogeny, and histology. Nevertheless, some questions have still not been clarified until today, e.g. the number of valid species. Based on the skull, all European findings until now are referred to as one species only (*Plateosaurus engelhardti* von Meyer, 1837). The Frick locality in Switzerland yielded over 20 skeletal specimens so far. They originate from a 12-metre thick bone-bed dinosaur layer which likely covers several million years. In our study, we use geometric morphometrics with a set of 24 landmarks to describe the morphology of 15 ilia from Frick. We performed principal component analyses and linear regressions to investigate shape variability. Shape was not significantly correlated with size, and no shape difference was observed among the layers of the Frick bone bed. However, two groups were clearly separated based on size. By investigating the ilium in more detail, we hope to find whether the differences are due to intraspecific variability, sexual dimorphism, or are an indicator of different species.

How to get to the bottom of annelid evolution?

Paul Kalke & Conrad Helm

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Annelids are a mesmerising group of invertebrates showing a wide range of forms, colours and shapes. As they inhabit nearly all conceivable biospheres, their adaptive structures, like their head appendages, show a huge range of evolutionary transformations ranging from feather-like crowns, countless buccal tentacles to the total reduction of them. To take a fresh look on this key feature of polychaetes evolution we use an integrative morphological approach including severe morphological methods as e.g. immunohistochemistry and confocal laser scanning microscopy, SEM, TEM, AZAN-histology μ CT and 3D-visualisations. Herein we want to discuss two possibilities to manage difficulties in data acquisition and visualisation. In this context, we will focus on methods to overcome the non-permeability of annelid cuticles for immuno-histochemical approaches and try to present an easy and open-access solution to compensate aligning and sectioning artefacts during ultrastructure and histology based 3D-visualisations. In this talk, we hope to provide some solutions and suggestions for common problems during morphological investigations. Likewise, we want to encourage you to open your mind for open source software solutions and the search for individual solutions.

Of tracts, lineages, and evolution: An atlas of the *Tribolium* brain

Max S. Farnworth, Bart Geurten, Gregor Bucher & Volker Hartenstein

Insect brains are complex organs that despite their small cell number cause equally complex behaviours. Each species has a brain adapted to specific environmental surroundings and for specific behaviours. The insect clade has diverged largely in its brain architecture reflecting such adaptations. Therefore, studying brain anatomy is essential to understand adaptive processes in insects.

I want to present our current work on the brain of *Tribolium castaneum*, the red flour beetle. While previous efforts have examined four classic brain areas, our approach goes further by taking the whole brain into account. Using a simple acetylated tubulin staining, we were able to determine compartments, fascicles, and tracts of the *Tribolium* brain at three different developmental stages. Moreover, the tracts, particularly visible in the freshly hatched larva, can be assigned to developmental neural lineages.

Using the large database in *Drosophila melanogaster* we are currently comparing the two species, in an – to our knowledge – unique way with respect to its completeness and simple methodology.

We hope that this work is a scaffold for future studies in *Tribolium*. Moreover, we think that this approach can be used in many insect species to generate a more complete understanding of brain anatomy and evolution.

A key developmental framework in the key annelid *Owenia fusiformis*

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Annelids are a group of segmented worms; whose embryos exhibit spiral cleavage and a variety of larval forms. While most modern embryological studies focus on species nested in Pleistoannelida, a few recent studies started to look into *Owenia fusiformis*, a member of the sister group to all remaining annelids to understand annelid. However, the early cleavage and morphogenetic events leading to the formation of the mitraria larva of *O. fusiformis* remain unexplored. Here we show that *O. fusiformis* undergoes equal spiral cleavage. Cleavage results in a coeloblastula approximately five hours post fertilisation (hpf). Gastrulation occurs via invagination and completes at 9 hpf, with putative mesodermal precursors and the chaetoblasts appearing 10 hpf. At 11 hpf, the apical tuft emerges, followed by the first *elav*⁺ and synaptotagmin⁺ neurons in the apical organ and the prototroch by 13 hpf, much earlier than previously recognized. Muscles develop around 18 hpf and by 22 hpf the mitraria swims. As the mitraria feeds, it grows in size and the prototroch expands through active proliferation. The larva becomes competent after ~3 weeks post fertilization. Altogether, our study identifies the major developmental events during *O. fusiformis* ontogeny, defining a conceptual framework for future investigations.

A single origin of excretory organs

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Most of the bilaterally symmetrical animals (Bilateria) excrete through specialized excretory organs, such as kidneys, metanephridia, and protonephridia. However, due to the morphological diversity of the excretory organs, it remains unknown whether those structures evolved from a common ancestral organ or appeared several times independently during animal evolution. In order to answer the question about the origin of excretory organs, we investigated the molecular pathways and structural genes involved in the development of diverse excretory organs in six animal species that represent the major evolutionary lineages of Bilateria (i.e. Spiralia, Ecdysozoa, and Deuterostomia). We show that excretory organs of phoronid, brachiopod, annelid, onychophoran, priapulid, and hemichordate express a set of highly conserved transcription factors. These results, combined with the data from planarians and vertebrate models, indicate the presence of an ancient molecular patterning of bilaterian excretory organs. Additionally, we showed that nephridia of phoronid, annelid, onychophoran and hemichordate express structural molecules that are also used for ultrafiltration by vertebrates and flatworms. Our results provide new evidence for the homology of all ultrafiltration-based excretory organs, which probably originated from a simple protonephridium-like structure in the deep geological past.

The stick insect attachment system: evolutionary ecology and functional morphology

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Insects have developed a broad array of specialized structures on their feet to properly attach to the vast diversity of natural surfaces. Particularly stick and leaf insects (Phasmatodea) witness a strong co-evolution with plants that caused such specializations on their tarsi. These include a wide range of functional surface microstructures on the attachment pads. We carried out broad scanning electron microscopy analyses of the attachment structures of phasmids and inferred the phylogenetic context based on molecular data. The distribution of the attachment microstructures among phasmids suggests a high degree of convergence of the structures, in accordance with their ecological relevance. The functionality of the attachment devices depends on different levels of structural organization including the microscale (contact mechanics at the interface with the substrate) as well as on the macroscale (tarsus compliance depending on the tarsal length). To access these two factors on stick insect attachment, we carried out behavioral observations and experiments, as well as force measurements on species with different tarsus morphology (i.e. attachment microstructures and tarsal length) exposed to substrates with different qualities (i.e. different surface roughness and substrate geometry). Our results revealed (1) an effect of the pad microstructures on the performance on different degrees of surface roughness and (2) a strong role of tarsal length on the performance on different macroshapes of the substrate.