

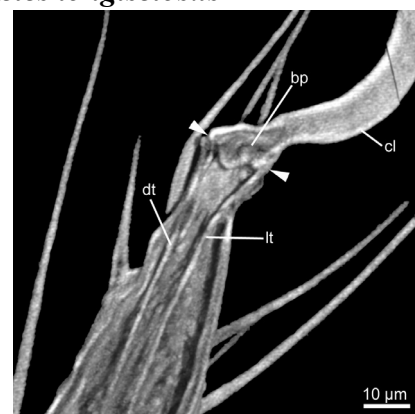
	Experiment title: Development of <i>Archezogzetes longisetosus</i> (Acari, Oribatida), a model organism for chelicerate evolution	Experiment number: SC-2127
Beamline: ID 19	Date of experiment: from: 09. Dec. 2006 to: 11. Dec. 2006	Date of report: 30. Nov. 2007
Shifts: 6	Local contact(s): Dr. Lukas Helfen	<i>Received at ESRF:</i>
Names and affiliations of applicants (* indicates experimentalists): Dr. Michael Heethoff, University of Tübingen Prof. Dr. Oliver Betz, University of Tübingen		

Report:

The experiment SC-2127, initially planned and approved with 9 shifts, was splitted in two separate experimental sessions with each 6 shifts. The first session was held in December, 2006, and used phase-enhanced tomography for the analyses of the functional morphology (muscles and cuticle) of different oribatid mite species and a fossil, 40 million year old specimen in Dominican amber (this report). The second session (separate report) was held in March 2007 and used quantitative phase tomography (holotomography) for the analyses of the developmental biology and differential imaging of soft tissues of *Archezogzetes longisetosus*.

Functional morphology and force production of tarsal claws of *Archezogzetes longisetosus*

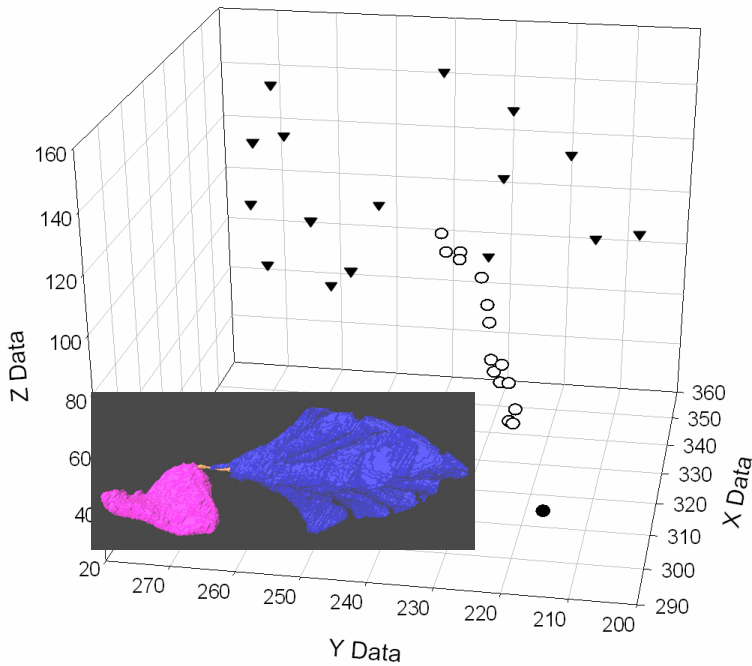
Due to their small body size (~0.8mm), oribatid mites (Acari, Oribatida) are only little studied with respect to their internal morphology and no studies were available which investigated functional aspects of morphological structures. We used phase enhanced tomography with an effective pixel resolution of 0.27 μ m to describe the functional morphology of locomotory appendages (see figure on the right showing the internal organisation of the claw articulation and the tendons connected to transmit muscle forces to the claw; bp: basilar piece, cl: claw, dt: depressor tendon, lt: levator tendon) and combined this information with experimental measurements of force production using micro-fore-transducers (Heethoff & Koerner, 2007). We showed that the oribatid mite *A. longisetosus* produces disproportionately high forces and, with respect to its body size, could be denoted as the strongest animal known so far.



Functional morphology of the feeding biology of *A. longisetosus*

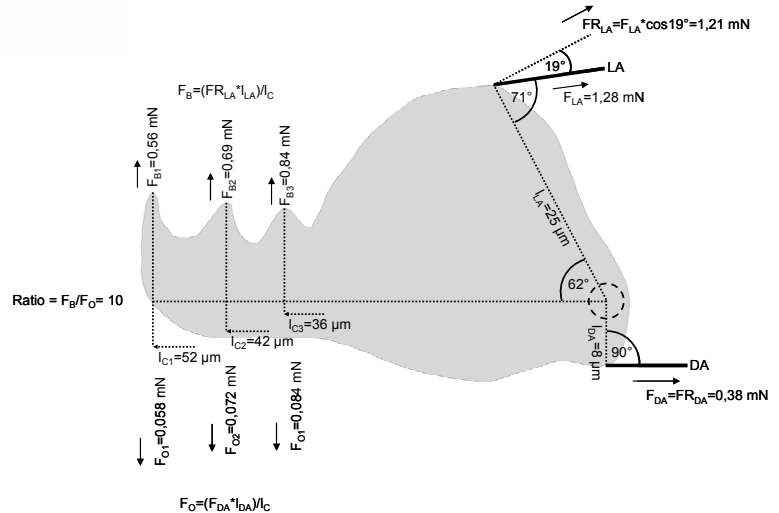
Oribatid mites are unique among chelicerate animals since they use particulate food with internal digestion instead of fluid food with external digestion. Coming along with this strategy, many physiological adaptations connected to the digestion of particulate food, were necessary.

Chelicerae: The chelicerae are the most important mouthparts in the Chelicerata and in most groups they are used to inject digestive fluids and/or poison in the prey. The use of particulate food added a completely different function to the chelicerae of oribatid mites: mastication. The high resolution of our data and the fact that the measurements are non-destructive, allowed me to build up the first functional model of a chelicera in general and even the first functional model of any microarthropod mouthparts. I included all muscles with their functional cross sectional areas, their effective angles to the force axis and their exact insertion points. The figure on the left shows the three-dimensional arrangement of muscle insertion points of the strong chelicera-closing-muscle and a reconstructed model of the muscles and the



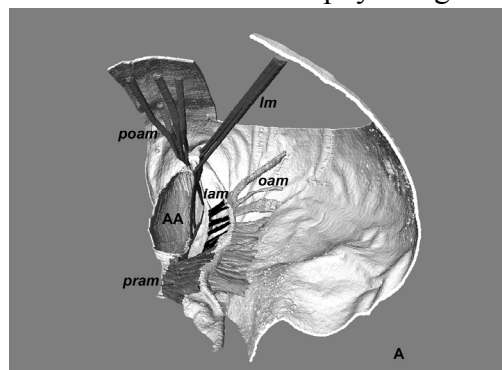
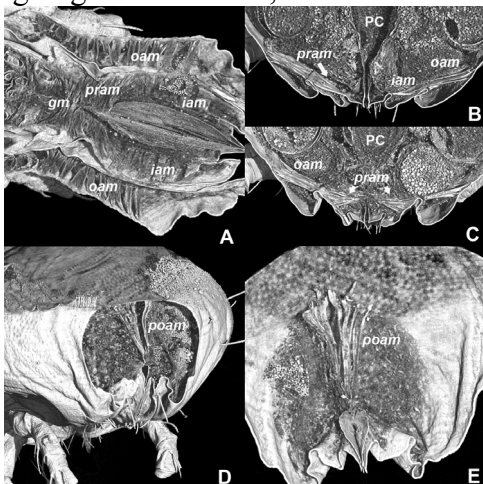
movable digit of the chelicera.

On the right side, I give a functional model of the movable digit of the chelicera with all angles and theoretically exerted forces.



Defecation: Highly interesting with respect to the functional morphology is the defecation process of oribatid mites. The use of particulate food leads to the production of extremely large faecal pellets, hence the anal opening needs to be able to release these large particles (Heethoff & Norton, submitted). A complex system of different muscle groups is involved in this process, and was described with contradicting suggestive functions in the past literature (cf. Betz et al., 2007). We used the phase contrast tomographic data (resolution: 7 μm) to analyse and describe all muscle groups in their natural state (left figure below showing different aspects of muscle organisation) and to build a functional model of all associated muscle groups.

The figure below on the right shows the functional model of all associated muscle groups (AA: anal atrium, gm: genital muscles, iam: inner anal muscles, lm: lateral muscles, oam: outer anal muscles, PC: postcolon, poam: post-anal muscles, pram: pre-anal muscles). Together with *in vivo* examinations of the defecation process we provided the first functional model of this physiological essential trait (Heethoff &



Norton, submitted). We showed that important muscle groups, previously described as dilators, are in fact functionally constrictors and vice versa.

Morphology of a fossil microarthropod in amber

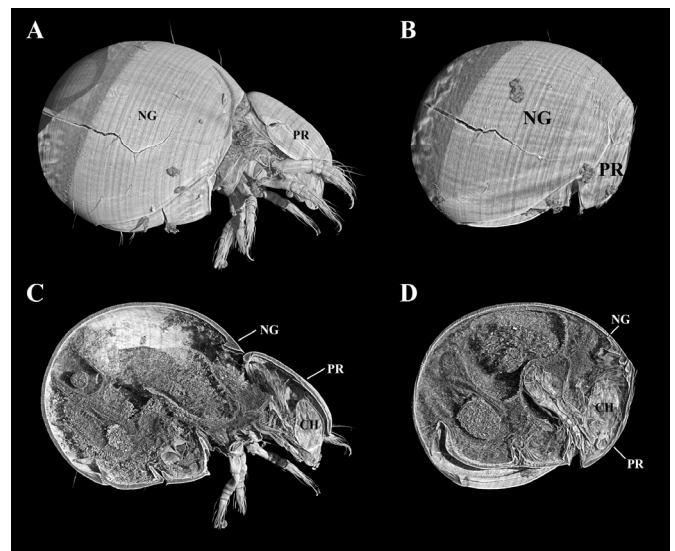
In the last years, some studies were published dealing with the non-destructive reconstruction of fossil materials, also in amber. However, most of the studies dealt with larger specimens or petrified samples with high absorption contrast. We conducted the first measurements using synchrotron X-ray microtomography to reconstruct the external and internal morphology of a 40 million year old specimen of an oribatid mite (family Neolioididae), deposited in Dominican amber. The left figure below shows a rendition of the external morphology. Species of this family carry the juvenile exuvial scalps on their notogaster and these were also



preserved. The preservation of external traits was good enough to identify and reconstruct taxonomically important characters, hence, we were able to describe a new (although extinct) species for the first time based on this method. The right figure is a sum-along-ray projection that shows the excellent preservation of external and internal cuticular elements.

Functional morphology of ptychoidy in oribatid mites

Besides a variety of chemical defensive mechanisms, oribatid mites display different and morphologically complex mechanical defensive adaptations such as large seta and cuticular shields. Some evolved the opportunity to enclose themselves completely to achieve a seed-like appearance (ptychoidy). We scanned different species of the group Euphthiracaroida for comparative and functional analyses of this important trait and showed that many of the adaptations are phylogenetically significant (Schmelzle et al., submitted). Data analysis is still in progress and the next step is the production of a functional model of ptychoidy which includes all muscles and cuticular elements. The figures on the right shows renditions of the external and internal morphology of *Phthiracarus globulosus* in an extended and encapsulated state.



Publications

Results from the first session were already published or are submitted for publication as follows:

Betz, O., U. Wegst, D. Weide, M. Heethoff, L. Helfen, W.-K. Lee & P. Cloetens (2007): Imaging applications of synchrotron x-ray micro-tomography in biological morphology and biomaterial science. I. General aspects of the technique and its advantages in the analysis of arthropod structure. - Journal of Microscopy. 22: 51 – 71

Heethoff, M. & L. Koerner (2007): Small but powerful: the oribatid mite *Archezogetes longisetosus* Aoki (Acari, Oribatida) produces disproportionately high forces. – The Journal of Experimental Biology **210**: 3036-3042

Heethoff, M. & R. A. Norton (2007): Functional morphology of idiosomatic muscles of *Archezogetes longisetosus* Aoki (Acari, Oribatida), with a focus on the defecation process. - Journal of Morphology, submitted

Schmelzle, S., R. A. Norton & M. Heethoff (2007): The ptychoid defensive mechanism in Euphthiracaroida (Acari: Oribatida): A comparison of exoskeletal elements. – Soil Organisms, submitted

Manuscripts of all other experiments mentioned above are about to be submitted.