ESRF	**Experiment title:** Investigation of insect morphology using hard X-ray microscopy and microtomography	**Experiment number:** LS-1575
Beamline: ID22	**Date of experiment:** from: 8.2.00, 9.6.00 to: 15.2.00, 13.6.00	**Date of report:** September 1, 2000
Shifts: 18	**Local contact(s):** Ch. Rau, T. Weitkamp	*Received at ESRF:*

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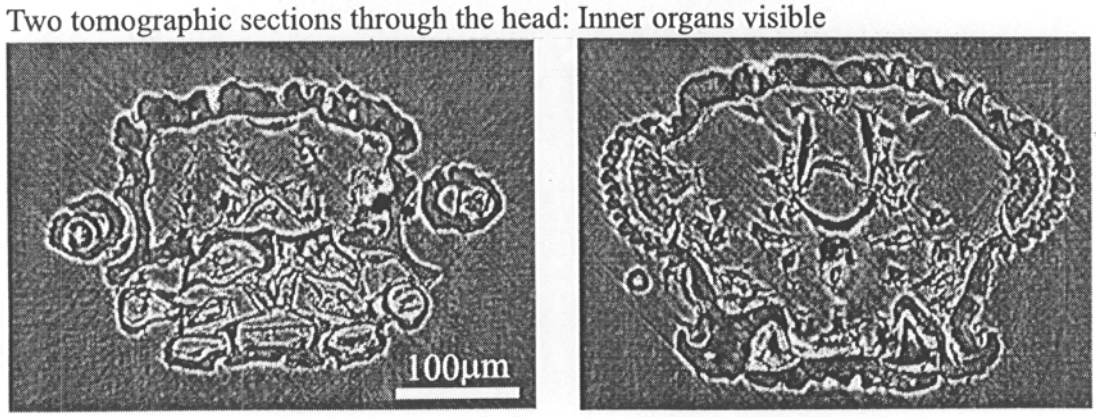
Report:

The morphology of various insects was investigated by phase contrast microtomography and hard x-ray full field microscopy using compound refractive lenses. For the first time, full field microscopy was combined with tomographic techniques to obtain the magnified tomogram of a sample.

At the ID22 microtomography station, high resolution phase contrast tomograms of the following samples were recorded: Head of *Eunicumus anthracinus*, head of *Eunicumus transversus*, full body of *Hoplopleura*, egg of *Hoplopleura*, leg of *Eunicumus tarsus*, front (*Pro*) and middle (*Meso*) leg of *Priacma serrata*, and the head of *Drosophila melanogaster*. Two selected reconstructed slices of *Eunicumus anthracinus* are shown in Fig. 1. Full 3D-reconstructions of all samples have been obtained.

1. Phase contrast tomogram of Eunicumus anthracinus
Two tomographic sections through the head: Inner organs visible

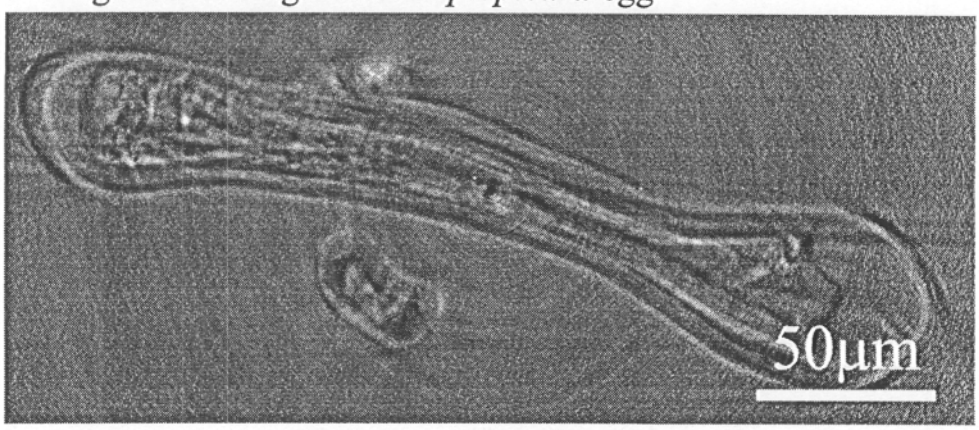


Experimental parameters: $E = 20.8$keV, FReLoN2000 (20x objective, 3.5μm LAG:Tb scintillator), sample detector distance 3-5mm.

Full field microscopy: Using compound refractive lenses (CRL) a full field microscope was built at ID22 during the experiment. Due to its large depth of field (several mm), micrographs of all investigated samples are sharp projection images. For the first time, full field microscopy was combined with tomographic techniques to obtain the 3-D structure of a samples. Since the resolution of hard x-ray micrographs is superior to ordinary projection images, magnified tomography can yield the 3-D structure with higher resolution. For smaller samples, such as *Hoplopleura*, high resolution is important for the phylogenetic evaluation. The limited field of view of the full field microscope (approx. 300μm) requires the samples to be small. The egg of *Hoplopleura* was an ideal sample for this technique.

A magnified tomogram (magn. 7.6) of the egg of *Hoplopleura* was recorded. 1250 enlarged projections over 180° were recorded at 27.5keV with an effective pixel size of 184nm (exposure time 15s per projection). The reconstruction of a slice is shown in Fig. 2. With a CRL (Al, $N = 60$, $f = 2420$mm) a resolution of 340nm is expected. However, due to spherical aberration (imperfections in the parabolic lenses used), the projection images did not reach this resolution. The reconstruction has a resolution of about 1μm.

2. Magnified tomogram of *Hoplopleura* egg



The results are currently being phylogenetically evaluated by T. Hörnschemeyer and G. Tröster at the institute for zoology at the University of Göttingen, Germany. A comparison of the method with conventional sectioning techniques will then be possible.

[1] B. Lengeler, *et al.*, "A microscope for hard x-rays based on parabolic compound refractive lenses," *Appl. Phys. Lett.*, **74** (26), 3924-3926 (1999)