ESRF	**Experiment title:** Relation between mechanical properties and structure of silkworm silk as a function of humidity content	**Experiment number:** SC-2464
Beamline: ID-13	**Date of Experiment:** from: 17.04.08 to: 21.04.08	**Date of Report:** 05.12.08
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Report:

Natural silks exhibit extraordinary mechanical properties, combining high tensile strength with a high elongation at failure. Producing man-made, synthetic polymer fibres with comparable properties (i.e. Kevlar) requires a considerable input of energy compared to natural silk fibres, which are spun from aqueous protein solution at ambient temperature. Similar to almost all biomaterials silk is hierarchically structured on many length-scales. On a mesocopic level silk is a semicrystalline nanocomposite with ordered regions (β-sheet fibroin nanocrystals) embedded in a softer matrix of disordered material [1]. The matrix is accessible to water, and the water content drastically influences the mechanical properties of silk fibres since it penetrates the disordered matrix but not the crystalline. The aim of our experiments was to establish a model for silk incorporating the macroscopic mechanical behaviour on the basis of morphology and molecular processes under influence of water amount in the material. To this end, we carried out *in situ* tensile tests during synchrotron radiation X-ray diffraction experiments in a controlled relative humidity (RH) atmosphere.

In the experiment reported here we used a new improved setup for stretching in an atmosphere with controlled humidity. Data analysis is still in progress; only preliminary results are reported here.

A piezo stretching device for the simultaneous acquisition of X-ray diffraction patterns and of force-elongation data of single fibres was developed for the experiment at ID13 (Fig. 1). Constant humidity is ensured by encapsulating the device with Kapton windows. The integrated basin was filled with silica gel (relative humidity 8 %) or water (RH 92 %). Additionally, a NaCl-solution was used to achive RH values of about 76%. The ambient relative humidity in the air-conditioned hutch was 42 % (open cell operation). We extracted single fibres with the help of fine tweezers in an optical microscope and glued them into small pre-fabricated plastic frames. Typical fibre length was 2-3 mm with a free length of 1.1 mm in the frames.

We used the so-called scanning setup of ID13 with a microbeam (about 1 μm diameter at the sample position) produced by KB mirrors. The whole experiment was monitored from the side with a high-resolution CCD Camera through a telecentric lens system. Comparison of two subsequent images allows for a determination of local strain. The tensile strain experiments were done with two kinds of strain exitation: 1. Slope strain, with constant strain rate of 0.025%/s. 2. Step-wise strain with 0.5 μm steps (corresponding to strain steps of 0.045%; see Fig. 2). In the second type of experiments the sample was allowed to relax for 10 s, and then a scan across the fibre (5 μm step size, 10 steps) was carried out. In addition to the usual horizontal scans, the diagonal scans along the line with angle of 45° was carried out. Acquisition time for a 2D diffraction pattern on was 1 s with 0.88 s between subsequent exposures.

The lattice strain of the β-sheet fibroin crystals in the direction of the tensile stress can be determined from the shift of the radial position of the 002 reflection (Fig. 2). The constant stress scenario has previously been suggested by Raman spectroscopic results [4]. It corresponds to a serial arrangement of crystalline and amorphous regions in the composite, known as the Reuss model in polymer physics. Water penetrates the disordered regions and softens the whole fibre, which consequently can be elongated further with less force.

Fig.1 Stretching setup with humidity control and sample observation telescope as installed on ID13.

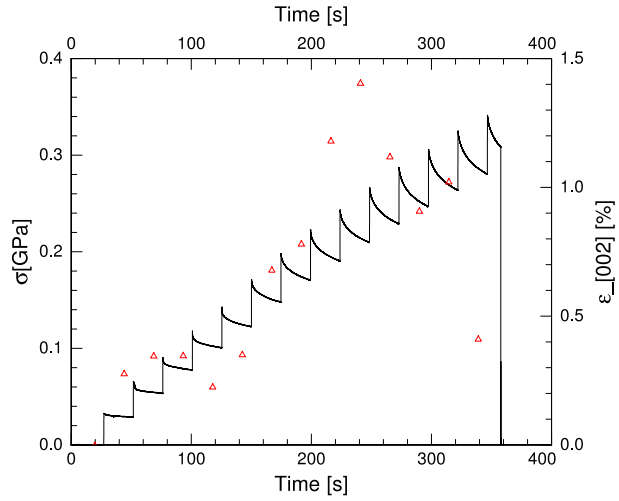

Fig.2 Force-time curve of a single silkworm silk fibre (10% RH) shown in black. The fibre breaks at a strain of max = 5.90%. Force relaxation on the steps is clearly visible. The red triangles show the crystal strain inside the microfibrils as calculated from the shift of the meridional fibroin 002 reflection position with increasing strain.

The deformation of the fibroin crystal is very important (up to 1.4 %) [3]. However, the experiment suffered from beam damage leading to premature rupture of the fibre. Future experiment will have to be carried out without time consuming scans across the fibre (secondary beam damage!) but rather with a constant strain rate and diffraction from different points on the sample.

The analysis of complementary relaxation experiments with high time resolution to probe the viscoelastic properties of silk is still in progress.

References

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