WAXS and SAXS on Invertebrate Jaws Revealed Copper Mineral Particles

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Introduction

In 1980, Brian and Gibbs discovered a biomaterial of very peculiar composition — the jaws of the marine polychaete worm Glycera, consisting of mainly protein and a few percent of copper. They were able to show that the high copper level was a characteristic feature of the jaws and suggested a structural function. However, it remained unclear in what form the copper was present in the jaw. The aim of the experiments described here was to find out if the copper was organized in clusters and if there was a crystalline phase.

Methods and Materials

Glycera jaws were used as dissected from the animal and glued to thin glass fibers for wide-angle and small-angle x-ray scattering (WAXS and SAXS). The samples were mounted on a motorized stage and scanned through the 100-μm-thick beam. The sample-to-detector distance for WAXS was 6 cm, and a wavelength of 1.24 Å was used. The setup for SAXS was essentially the same, only the sample-to-detector distance was increased to 525 cm. Thus, SAXS and WAXS could be done on the same samples. In order to distinguish the SAXS signal from pores and microcracks from the signals due to nanostructural features like copper clusters, the scattering contrast was varied by measuring dry and wet samples. Wet samples were encapsulated in a Kapton® bag to prevent them from drying during the experiment.

Results and Discussion

The position-resolved WAXS measurements showed that there is a crystalline phase within the last 200-300 μm next to the very tip of the jaw (Fig. 1a). The tip of the jaw is also where the highest levels of copper have been found. The experimental diffraction pattern was compared with known copper minerals from the literature, and an excellent match was obtained with the calculated pattern of the copper- and chlorine-containing mineral atacamite ($Cu_2(OH)_3Cl$) (Fig. 1b). This result is further supported by the finding of a high spatial correlation of the copper and chlorine in the sample obtained by microprobe analysis done at the University of California, Santa Barbara. This finding is remarkable in that this is the first time that a copper mineral was found in a bioorganism.

The corresponding SAXS patterns (Fig. 2a) showed a strong, highly anisotropic scattering signal at the tip of the jaw that changed little upon hydration. We thus assume that the anisotropic signal from the tip is due to the strong electron density contrast between the mineral particles and the protein and that it is unaffected by hydration. The anisotropic pattern was always oriented perpendicular to the outer shape of the sample, indicating a strongly preferred orientation of the mineral particles with their longitudinal axis parallel to the sharp tip. In the rest of the jaw, the signal was isotropic and much weaker — almost vanished in the wet jaw — and therefore most likely a result of the pores. Since the region where the crystalline phase occurred corresponds with the region showing the strong SAXS signal, we propose a crystal arrangement, as shown on the left in Fig. 2.

Acknowledgments

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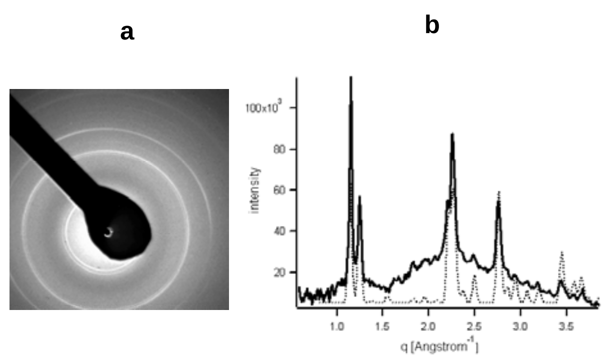

FIG. 1. (a) X-ray diffraction pattern measured at the tip of a Glycera jaw. (b) The resulting pattern (full line) matched almost perfectly the calculated pattern for atacamite (dashed line).

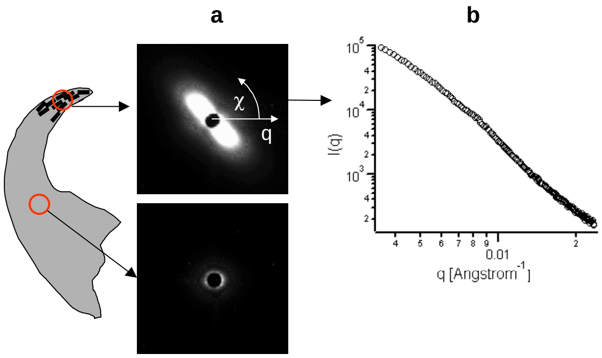

FIG. 2. (a) SAXS patterns measured at the tip and the middle, respectively, of a wet jaw. The red circles on the left indicate the beam size. (b) Radially averaged intensity obtained from (a). The sketch on the left shows a model of the arrangement of the mineral crystallites in the Glycera jaw.

Reference

[1] P. E. Gibbs and G. W. Bryan, J. Marine Bio. Assoc. U.K. **60**, 205-214 (1980).