Sectioning Fish Lenses with ROWIAK TissueSurgeon: Artifact-Free Sectioning with Femtosecond Laser Technology

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Introduction

The preparation of histological sections of fish lenses with common microtomy is a very critical process as the material tends to crack (Fig. 1). Common microtomes put pressure and shear forces on the material, which is very brittle, even if embedded. Several methods of embedding were tested. The aim of the present study is to develop a procedure to section fish lenses without these common artifacts.

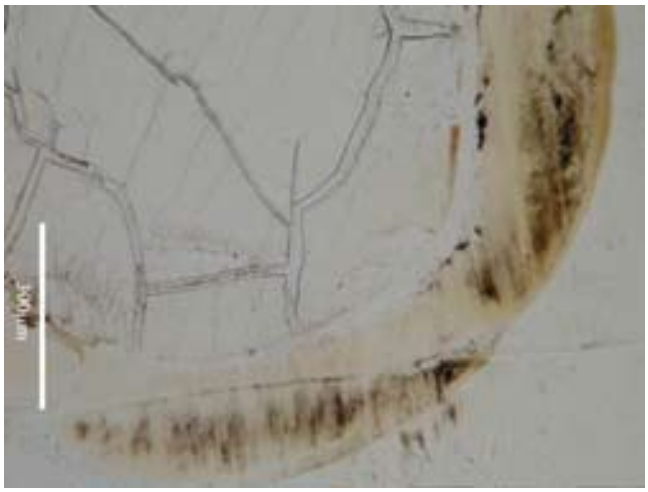

Fig. 1: Histologic image of a fish lens sectioned with a common microtome. Note the extensive cracking.

Material and Methods

Fish lenses were stained red for orientation and embedded into Epon. Embedded blocks were trimmed with a diamond saw to remove excess embedding medium. The surface was mounted on a microscope slide with Technovit 7210 (Heraeus Kulzer) and cured for 30 min. Sections were prepared with ROWIAK TissueSurgeon at 20 µm thickness. Sectioning was monitored with the OCT-device of the TissueSurgeon (Fig. 2). The sections were examined with a Zeiss Axioskop microscope in transmitted light and phase contrast.

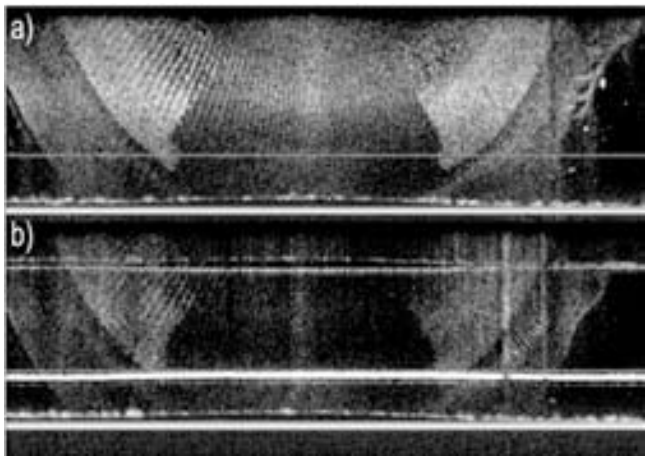

Fig. 2: OCT-image of a fish lens a) before sectioning, b) after sectioning

Results

Sectioning of fish lenses with the ROWIAK TissueSurgeon is successful. Lens sections are free of cracks and surface artifacts (Fig. 3a). The structure of the lens can be examined in phase contrast (Fig. 3b). Rows of cells, rich in transparent proteins, can be observed.

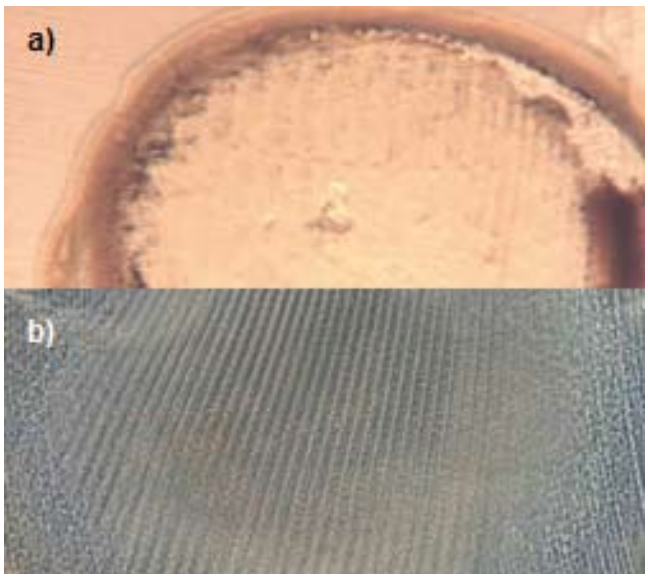

Fig. 3: Sections of fish lenses prepared with the ROWIAK TissueSurgeon (a) bright field, (b) phase contrast

Conclusion

Laser Microtomy is a novel method permitting for the first time sectioning of embedded brittle tissues, such as hard fish lenses, without artifacts. The subject of current development is to section native fish lenses.