Three-dimensional X-ray microscopy of zebra sh larvae

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Successful tomographic imaging of soft tissues with micrometer resolution includes preparation, acquisition, re- construction, and data evaluation. Tissue preparation is essential for hard X-ray microtomography, because staining- and embedding materials can substantially alter the biological tissue post mortem. We performed to- mographic imaging of zebrafish embryos in alcohol and after paraffin embedding with a conventional X-ray source and at a synchrotron radiation facility. The resulting multi-modal, three-dimensional data were registered for direct comparison. Single-cell precision was reached for the synchrotron radiation-based approach, which allows for segmentation of full organs such as the embryonic kidneys. While this approach offers an order of magnitude higher spatial resolution, many of the anatomical features can be readily recognized with the more accessible laboratory system. Propagation-based data acquisition enabled us to demonstrate the complementary nature of the edge-enhanced absorption contrast- and the phase contrast-based modality for visualizing multiple microanatomical features. While ethanol and paraffin embeddings allowed for identification of the same anatomical structures, paraffin-embedding, however, led to more artefacts and shrinkage. The presented multi-modal imaging approaches can be further extended to visualize three to four orders of magnitude larger volumes such as adult zebrafish or complete organs of larger animals such as mouse brains. Going towards even larger volumes, such as the human brain, presents further challenges related to paraffin embedding, data acquisition and handling of the peta-byte scale data volumes. This study provided a multi-modal imaging strategy by the combination of X-ray sources and sample embeddings which can play a role in addressing these challenges.

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