

## BioXRM: Microtomography of embryos, tissues, cells, and molecules

3D imaging has become a familiar player in developmental and comparative biology, offering ever more realistic views of the native structures of organisms and materials. For morphology-based studies of development and evolution, X-ray microtomography (microCT) is the most suitable method to visualize 3D micromorphology in whole embryos and other intact samples. Contrast-enhanced microCT can produce images with microscopic detail and high contrast among various non-mineralized tissues – histology without sectioning.

The accuracy of any analysis based on 3D images depends first on the quality of the sample, and second on the fidelity of the imaging process. Thus we are refining our methods for fixation, tissue stabilization, and contrast staining of whole samples for microCT imaging. The images generated by microCT are size-calibrated and suitable for quantitative 3D analyses of developmental morphology. We are currently establishing datasets and workflows to measure and model intraspecific variation, asymmetry, and growth during development.

Ongoing work on dual-energy (spectrally-sensitive, “two-color”) microCT has demonstrated simultaneous imaging of different tissues or materials, e.g. skeletal hydroxyapatite amid counterstained soft tissues, and selective labeling and imaging of melanocytes (pigment cells) in whole animals. We are extending this work to 3D localization and quantification of molecular probes in counterstained whole embryos. Further collaborative work is aimed at imaging cultured and regenerating tissues within artificial scaffolds and incorporating novel biomarkers such as nanoparticles.

Digital volume images are inherently shareable, and we have recently published the first new (millipede) species description to be based partly on its cybertype – a set of virtual specimens made from microCT images of the physical type material for the species. We have also made a developmental atlas of the squid *Euprymna scolopes* available online, and we are currently creating a high-resolution 3D atlas of Sprague-Dawley rat development as a basis for analyzing mutations, experimental perturbations, and for quantitative comparisons with other species.

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