RAPID COMBINED LIGHT AND ELECTRON MICROSCOPY ON LARGE FROZEN BIOLOGICAL SAMPLES

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The use of large unfixed frozen tissue samples (10x10x5 mm^3) for combined light microscopy (LM) and electron microscopy (EM) is described. First, cryostat sections are applied for various LM histochemical approaches including in situ hybridization, immunohistochemistry and metabolic mapping (enzyme histochemistry). When EM inspection is needed, the tissue blocks that were used for cryostat sectioning and are stored at -80°C, are then fixed at 4°C with glutaraldehyde/paraformaldehyde and prepared for EM according to standard procedures. Ultrastructurally, most morphological aspects of normal and pathological tissue are retained whereas cryostat sectioning at -25°C does not have serious damaging effects on the ultrastructure. This approach allows simple and rapid combined LM and EM of relatively large tissue specimens with acceptable ultrastructure. Its use is demonstrated with the elucidation of transdifferentiated mouse stromal elements in human pancreatic adeno-carcinoma explants grown subcutaneously in nude mice. Combined LM and EM analysis revealed that these elements resemble cartilage showing enchondral mineralization and aberrant muscle fibres with characteristics of skeletal muscle cells.

References

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