Core facility Cellular Imaging: Electron Microscopy Centre Amsterdam
Technique; TEM, resin embedded

Introduction techniques:

This technique is broadly applied in cell biological research and diagnostic pathology. Biological (patient) material is fixed, dehydrated and stained with heavy metals (lead, uranium, osmium). After embedding in plastic resin ultrathin sections are cut on a diamond knife and materials are analyzed using the TEM. With the electron dense counter- staining (positive staining) you can obtain an ultrasharp image with high contrast.

Specific counter-stains highlight different organelles and structures;

- Osmium — lipids, protein
- Lead — (lipo)proteins, glycogen, RNA
- Uranyl acetate — DNA(chromatin), ribosomes, protein
- PTA tungsten — polysaccharides, glycoprotein, collagen

Sample preparation

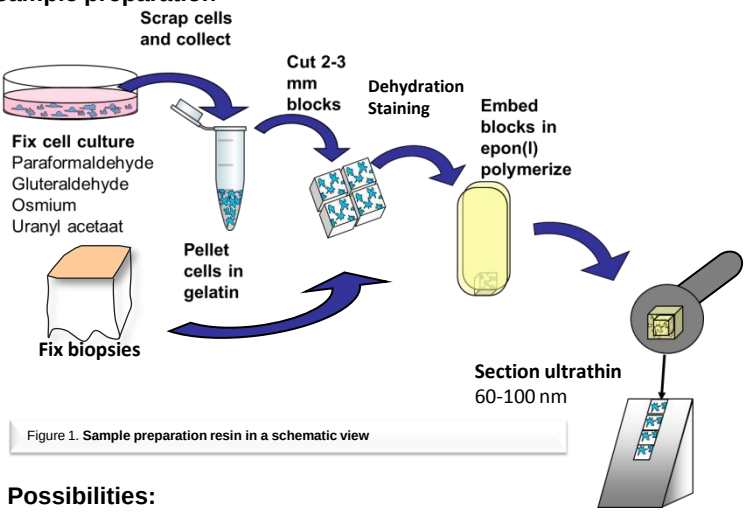


Figure 1. **Sample preparation resin in a schematic view**

Possibilities:

Morphological analysis of tissues at cellular and subcellular resolution. Ultrastructural changes within cells, cell organelles.
Fast and suitable for almost all biological samples;

- Bone
- All Organs; experience in spleen, kidney, muscle, liver, eye, skin
- Cell cultures, adherent and suspension cells
- Transwell and TC culture plates
- Bacteria in culture or cells

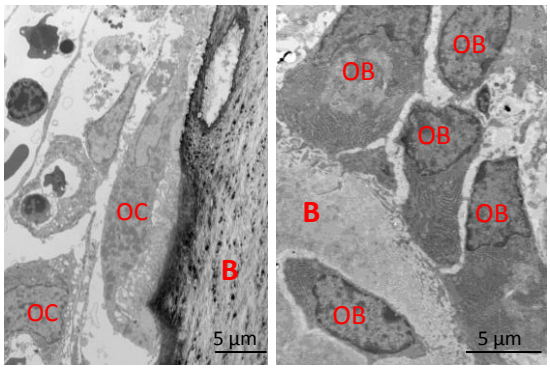
Limitations:

Resolution limited to 2 nm due to use of heavy metals
Sectioning needed, 60-100 nm.
Small sample size 1x1 cm^2

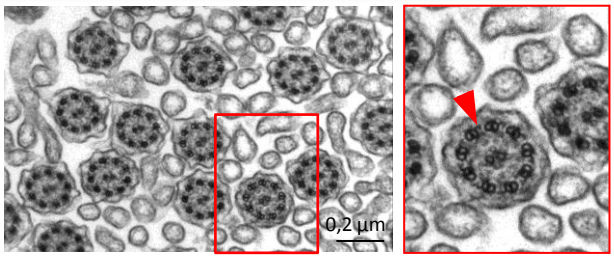
Needed:

Paraformaldehyde, glutaraldehyde fixed tissue or cultured cells (minimal 1 million cells). Cells will be dehydrated, stained and embedded in epon.

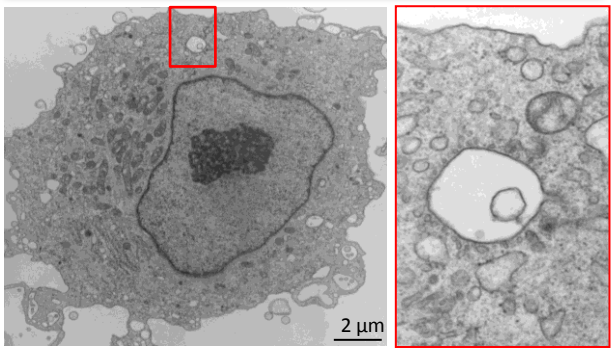
Examples results of the technique



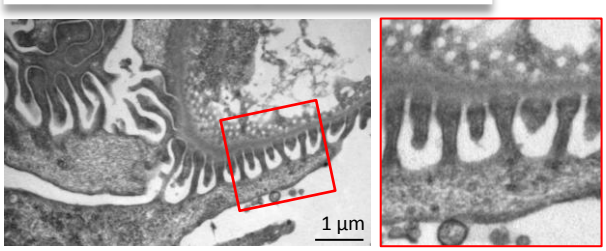
Osteoclast (OC) resorbing bone (B) (left) and osteoblast (OB) (right) generating bone material



Normal cilia morphology with dynein arms (arrow) in nasal epithelium (M. v.d. Bergh Weerman)



Langerhans Cell (MUTZ cell line) (W. Tigchelaar, C. Ribeiro, T. Geitenbeek)



Podocytes in the urinary space of the Bowman's capsule (M. v.d. Bergh Weerman)

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