TITLE: Phosphotungstic acid (PTA) staining on EM grids

PURPOSE: To demonstrate collagen (normal or abnormal) by EM, useful in the
diagnosis of Nail-Patella syndrome, collagenofibrotic glomerulopathy

SPECIMEN: Unstained ultrathin plastic section on EM grids

MATERIAL: 2.5% Phosphotungstic acid solution in 0.5-1.0 N Hcl.
1% periodic acid

EQUIPMENT: Usual staining apparatus for EM grids

METHOD: 1. Treat osmium-fixed tissue with period acid for 30 minutes at room
temperature
2. Wash
3. Stain grids for 15-30 minutes with PTA solution at room
temperature
4. Wash
5. View in the transmission electron microscope as black staining
localizing within collagen fibers

MICROSCOPE: Electron microscope

NOTE: - Osmium fixation may interfere with staining
- Pretreatment with periodic acid will help

REFERENCE: Pease DC, Bouteille M. The tridimensional ultrastructure of
native collagenous fibrils, cytochemical evidence for a carbohydrate

Courtesy of Surya V. Seshan, Weill Cornell Medical College, New York