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## Procedure For Using BUTVAR B-98

BUTVAR B-98 support films are mechanically stable, electron-transparent, and have minimum intrinsic structure. Their hydrophilic nature facilitates the spreading of the sections allowing even staining on the surface. This film can resist an intense electron beam during focusing and photography at high magnifications, or with the beam at cross-over. The mechanical stability is appreciated when mounting freeze-fractured replicas or serial sections, especially when slotted grids are used.

## **Procedure**

Stock solution - 0.25% BUTVAR (w/v) in chloroform:

- 1. Due to limited solubility the solution is unstable and sensitive to humidity. Heat (in a water bath) to 50°C with continual stirring and cool to 40°C 42°C just before coating grids. This heating procedure is extremely important to insure complete solubility of the BUTVAR and to obtain a good surface uniformity of the film free from irregularities, holes or wrinkles.
- 2. Use a pre-cleaned, 1x3 microscope slide (washed in 70% alcohol and flame dried).
- 3. Dip slides into stock solution and drain them in a vertical, inclined position in a dust free place (drawer or cabinet) on a paper towel or any absorbent material.
- 4. The upwards facing side is the thinnest (40nm). Dried slides are scored on the corners, and the film released by flotation into a staining dish filled with distilled water. Grids are arranged in 2 or 3 rows.
- 5. The film is retrieved with parafilm (2x4 inch) by gently touching one edge of the resin film, lowering into complete contact, then lifting up with a fast motion.

Coated grids can be used immediately and may be stored for several years.

Films for slotted grids are prepared in the same way. If grids are difficult to remove, the film may be scored around the periphery with the tip of a forcep, or the concentration of the stock solution can be reduced to 0.20%.

The stock solution is useable indefinitely when stored in an amber bottle, well capped, at room temperature.