

### Comments Re. Dr. Lederberg's Questions

Cronar is a selected grade of mylar.

Mylar is a polyester film - made of polyethylene terephthalate - the polymer formed by the condensation reaction between ethylene glycol and terephthalic acid.

The specific film which we are using is DuPont Type P40A "Cronar" plain motion picture leader, 16 mm wide, and perforated on two sides.

The material has an anti-static treatment on one side.

As we use the film, only one side is satisfactory for our purposes. To determine which side is "good" we apply the ethyl violet stain to both surfaces; the side that does not take up the stain is the side on which we impact our sample.

### Re. the Selection of Ethyl Violet

This dye was selected empirically. Our first thought was to detect the bacteria by their UV absorbing properties. This fizzled when we noted that the absorbancy at 265 m $\mu$  decreased quite sharply as the organism aged. A 14-18 hr. culture showed quite weak absorption of UV.

The next step was to look for a UV absorbing dye which would "enhance" the absorption of UV by the organism. After studying about 40 dyes, safranin-0 seemed most promising.

Then we looked at the visible spectrum. A study of 63 dyes showed ethyl violet to be the best. (Acid fuchsin is best for spores). Since the ethyl violet was better than the UV absorbing safranin-0, we chose the former.

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