AN IMPROVED METHOD OF STUDYING POLLEN TUBES THROUGH THE PREPARATION OF PERMANENT STYLAR WHOLE MOUNTS[1]

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For studies of pollen tubes of *Datura,* Buchholz (1931) devised a method of preparing temporary stylar whole mounts in lactic acid. Eigsti (1937) introduced a procedure for the preparation of permanent pollen-tube slides from these lactic acid mounts through the use of dioxan vapor for changing reagents. Eigsti's method, although satisfactory for small materials and stylar mounts of the *Liliaceae,* is not adaptable for styles of the *Solanaceae* because the specimens are often damaged when the cover glass is removed from the lactic acid mount. To overcome this difficulty the following method has been devised for the infiltration of the stylar tissue of *Nicotiana tabacum* with balsam without the removal of the cover glass.

1. A paper clip is used to hold the cover glass to the slide during the changes of reagents.

2. The temporary lactic acid mount is placed in a staining stender filled with 100 percent alcohol and kept there for two days. This procedure repeated three times insures replacement of lactic acid with 100 percent alcohol.

3. The slide is then transferred to a stender containing xylol. Three successive transfers through xylol at two-day intervals completely replaces the 100 percent alcohol with pure xylol. To achieve satisfactory results this exchange of reagents must be complete.

4. The slide is then transferred to a stender containing Canada balsam in xylol and kept in this stender for two or three days. It is removed from the balsam to an empty stender for drainage of the excess balsam which requires about one day. A reservoir for the excess balsam is created by raising the slide with a small glass rod placed in the bottom of the stender.

5. The slide is removed from the drainage stender, the paper clips are removed carefully, and the slide is then left to dry for one week prior to cleaning and permanent labeling.

6. Cleaning must be done with a cloth moistened slightly with xylol. Excessive pressure on slide or cover glass should be avoided during cleaning.

No destaining, shrinkage, or collapse of tissues has been noted in these slides, which have been on permanent file for more than one year. The entire pollen-tube population remains undisturbed during the process of making slides permanent by this method. Very few slides are damaged by mechanical injury during these manipulations.

Recently this method has been used satisfactorily in the preparation of permanent slides of fern gametophytes and fresh water green algae.

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[1]Contribution from the Botanical Laboratory of the University of Oklahoma, n.s. 68.

LITERATURE CITED

Buchholz, J. T. 1931. The dissection, staining and mounting of styles in the study of pollen-tube distribution. Stain Tech. 6: 13-24.

Eigsti, O. J. 1937. Permanent pollen-tube slides with the vapor method of changing reagents and dehydration. Stain Tech. 12: 53-54.