A new method for rapid mounting of monitor sections of plant material embedded in Spurr's resin

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Thick (2–8 μm) sections of plant material embedded in Spurr's epoxy resin can be affixed to clean microscope slides coated with chrome alum-gelatin. Sections are flattened and brought into close contact with the adhesive by rubbing firmly on filter paper placed directly over the sections. After staining with aqueous Toluidine blue or Azure/Methylene blue, sections are dried by blotting again with filter paper. No heat need be applied during mounting or staining and wrinkling of sections is thus avoided. Loss of sections during staining is minimized by using aqueous stains. Sections are dried and can be mounted in immersion oil or unpolymerized Spurr's medium within one minute of preparation.
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Dik (2–8 μm) sneë van plantmateriaal wat in Spurr se epoksie-hars ingebed is, kan op skoon voorwerpglasies bevestig word met chroomaluin-gelatien. Sneë word uitgestryk en in noue kontak gebring met die gelatienlagie deur hard op 'n laag filtreerpapier wat direk op die sneë lê, te vryf. Na kleuring met waterige Toluidienblou of Asuur/Metileenblou, word die sneë gedroog deur dit met filtreerpapier te klad. Geen hitte is nodig tydens kleuring of montering nie, dus word rimpeling van die sneë vermy. Verlies van sneë tydens kleuring word verminder deur slegs waterige kleurstofoplossings te gebruik. Sneë is binne een minuut na voorbereiding droog genoeg om in immersie-olie of ongepolimeriseerde Spurr-medium gemonteer te word.
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Introduction

The cutting of thick (2–8 μm) survey sections of plant material embedded in Spurr's epoxy resin (Spurr 1969) is widely practised in plant microtechnique. These sections are either used to check the orientation of the embedded material, or, more usually, to ascertain whether the sectioning plane has approached the area of interest. As this monitoring process may be repeated often, the mounting and staining of the sections must take up as short a time as possible.

Traditional methods of mounting such thick sections include the use of adhesives such as the standard mixtures of Haupt (1930) or Mayer, the subbing mixture of Pappas (1971) or diluted subbing (Warmke & Lee 1976) and gelatin in water (Grimley *et al.* 1965; Kosakai 1973). Sections are often dried down on slides without adhesive from water or 5% or 10% acetone (Alsop 1974) with subsequent heating (Spurlock *et al.* 1966; Hoefert 1968). Thorough drying of the slides before staining is usually essential (Trump *et al.* 1961; Shires *et al.* 1969). The drying process is usually heat assisted and may be protracted (Warmke & Lee 1976). Mounting of sections on partly polymerized epoxy resin (Braak 1977) is also slow, although good results are obtained.

Injudicious heating of sections is often the cause of wrinkling or corrugation of sections. This is especially the case when thick-walled plant material is handled because the epoxy resin expands during heating while the thick cell walls may not do so to the same extent. Although a method whereby sections may be dried down without wrinkling has been described by Harris (1978), the procedure takes at least 30 min before the sections can be stained.

As most of the described procedures are generally time consuming, the usual way in which such monitor sections are handled, is by floating a section on a drop of water or 10% acetone on a clean slide, which is then dried on a slide warmer or over a spirit lamp. The slide is then usually heated to bake the sections onto the slide. Such monitor sections are usually stained with Paragon-1301 (Spurlock *et al.* 1966) or with Azure II/Methylene blue (Richardson *et al.* 1960). Although the method is fast, a large proportion of the sections are sometimes lost during staining. The sections are nearly always badly wrinkled. Wrinkles which appear during staining can be prevented by the use of the method of Sommer *et al.* (1979), but this entails

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unacceptably long staining periods of 12–24 h.

A rapid method is described by which sections may be affixed to slides without wrinkles appearing and with enough tenacity to prevent all but very occasional section loss. This method has been used in this laboratory for some time.

Procedure

Sections (2–8 μm thick) of Spurr-embedded plant material are cut with dry glass knives and transferred with tweezers to drops of 10% acetone on alcohol-cleaned slides, previously coated with subbing mixture (Pappas 1971). Serial sections may be mounted in up to four rows, each section on its own drop of 10% acetone. A piece of Whatman no. 1 filter paper is placed over the slide on the drops containing the sections. The paper is held in position on the slide by pressing on it at one end. The sections are then fixed to the slide by rubbing the paper over the sections thoroughly with a finger nail. The free end of the filter paper is lifted, keeping it in position on the slide by pressure on the other end. Occasionally a few sections may adhere to the filter paper. These may be transferred to the slide, in their correct positions, by further rubbing after replacement of the paper on the slide.

The dry, flattened sections can be stained with Azure II/Methylene blue solution (Richardson *et al.* 1960), or Toluidine blue (Trump *et al.* 1961) for 30 s and the slide cautiously washed with water. Stains such as Paragon-1301 which contain alcohol cause unacceptable loss of sections during staining and washing. The sections adhering to the slide are dried by further rubbing with interposed filter paper as previously mentioned. These sections are dry enough to be covered immediately with immersion oil or unpolymerized Spurr's resin under a cover slip.

By following the above procedure, it is possible to mount, stain and examine the monitor sections within a relatively short period of time.

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