formed implies very selective deposition, as mentioned by Wardrop and Bland[3]. The foregoing staining reaction serves also as an excellent technique for clearly defining the size and shape of the pit membrane and it has been suggested that this technique should have general application in anatomical work.

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A Sealing Compound for use in Biological Work

In the course of rhizosphere studies, it was desired to measure continuously the respiration of intact sterile roots during the growth and development of a variety of plants. To separate the photosynthetic and respiratory activities of the portions above ground from the respiration of the roots, a material was required to prevent diffusion of gases from one zone to the other. The material had to fulfil several requirements in order to be effective: (1) be impermeable to gases and water; (2) be non-toxic to plants; (3) cause no physical damage to plants; (4) be sufficiently pliable to permit normal enlargement of stems; (5) withstand a continuous positive pressure of 0·5–1·0 lb./in.^{2}; (6) resist microbial and environmental deterioration; (7) be autoclavable; (8) be easily handled and applied; (9) be relatively inexpensive.

Although a variety of substances was tested, only one compound has been found which fulfilled all these requirements. This material is a viscous silicone fluid which solidifies at room temperatures on the addition of a catalyst. (Material is marketed by the Dow Corning Corporation, Midland, Michigan, under the trade name of 'Silastic *RTV*' (room temperature vulcanizing silicone rubber), and by the General Electric Co., Silicone Products Department, Waterford, New York, under the name of 'Silicone Rubber *RTV*'.) The time for complete vulcanization can be controlled by the type and amount of catalyst used; working time can be adjusted from 10 min. to several days. The plasticity can be increased by the addition of a silicone thinner (Dow Corning '200 Fluid', 20 centistokes). The base, catalyst, and thinner may be autoclaved separately and mixed aseptically just prior to pouring around the stem.

The rubber-like solid forms an impermeable seal with glass, metal, certain plastics, sand, and with plant roots and stems. Although a continuous positive pressure of only 0·5–1·0 lb./in.^{2} is regularly used in our work, the material is capable of withstanding considerably higher pressures, both positive and negative. A layer 10 mm. thick and 38 mm. in diameter withstood a continuous positive pressure of 7 lb./in.^{2} and a continuous negative pressure of 500 mm. mercury; a layer 10 mm. thick and 70 mm. in diameter withstood a pressure of 2 lb./in.^{2}. At higher pressures, the sealer itself does not leak, but the interface bond between the sealer and either the wall of the glass tube or the plant stem is ruptured.

Although a gas-tight seal is formed around the stem of plants, the seal is sufficiently pliable to permit normal development of the stem as well as of the entire plant. The consistency of the vulcanized product does not change during autoclaving nor during prolonged use, and may be employed at temperatures ranging from −70 to 500° F. Other physical and chemical properties are described by the manufacturers.

In our work, surface sterilized seeds are planted in short glass growth tubes (38 mm. internal diameter) contained in specially designed culture units, which will be described fully elsewhere. The roots grow into a sterile respiration chamber, and the upper portions of the plant are allowed to develop in air. To separate the two zones, the sealer is poured around the plant stem inside the growth tubes and allowed to solidify before a continuous air-stream is passed through the root respiration chamber. Both monocotyledonous and dicotyledonous plants have been successfully cultured in the presence of this material: legumes and corn have produced fruit; cotton, crucifers, cucurbits, and seeded bananas are at present growing vigorously.

Other compounds tested as sealers were not suitable for several reasons. Various combinations of paraffin, petrolatum and lanolin, as well as some carboxyvinyl polymers, were extruded from the growth tubes when pressure was applied; this difficulty was accentuated when the temperature of the environment was high. Several materials, for example, latex, poly*iso*butylenes, urethane polymers, polyacrylic and polyvinyl resins, were toxic; some set too rapidly for easy handling; some, for example, urethane polymers and some microcrystalline waxes, cured to a consistency that constricted the stem and prevented normal development; some, for example, various paraffin-petrolatum mixtures, butyl-latex emulsions, distilled acetylated monoglycerides, carboxyvinyl polymers, shrank on curing and failed to form a gas-tight interface with either the glass tube or the plant stem; some did not withstand autoclaving; and others, such as the polyethylene glycols, being water-soluble, were dissolved by condensation, transpiration and guttation water.

In addition to being used in sterile root cultures, the silicone material is also being employed in these Laboratories to seal surface-sterilized intact roots in tubes containing root-infecting organisms (Halmos, S., and Beckman, C. H., unpublished results); to make gas-tight gaskets for attaching respiration and transpiration chambers to leaves[1]; to make impressions of leaf surfaces for microscopic examination (Brun, W. A., unpublished results; and Zelitch, I., unpublished results); and to make closures for odd-shaped and hard-to-reach openings which pre-manufactured rubber stoppers do not fit. Additional applications are described by the manufacturers.

The non-toxicity of this compound to plants, as well as its other properties, should make it useful in a variety of biological studies.

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