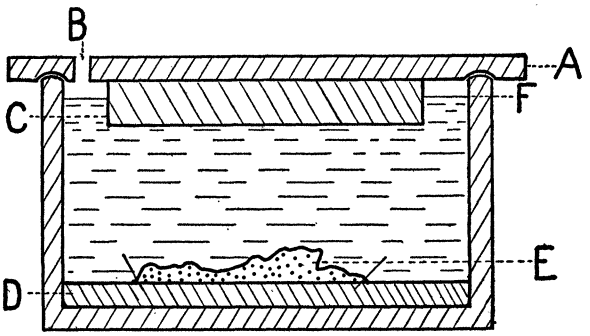
SCIENTIFIC APPARATUS AND LABORATORY METHODS

A CLEAR-VIEW SPECIMEN DISH

AFTER a number of experiments a museum dish has been evolved which can be used under the binocular microscope to good advantage. Valuable embryos and delicate dissections can be kept unharmed and viewed without optical distortion by this method. The accompanying figure gives the details of the dish.



A. Lid of stender dish.
B. Drilled hole for filling.
C. Plate glass disc cemented to A with Valspar.
D. Layer of wax.
E. Specimen attached to wax.
F. Upper surface of preserving fluid.

Clear glass imported stender dishes of various sizes were obtained. These had plate glass covers with a groove ground on the under surface. We next secured thick discs of clear polished plate glass (C in figure), somewhat smaller in diameter than the inner diameter of the stender dish. These discs were cut and polished at a local mirror and windshield factory. Such a disc (C) was then cemented to the under side of each lid (A). After many trials, an ideal cementing substance was found to be clear Valspar. The method for cementing was as follows: lid (A) is put with the upper surface down on a warming plate. A number of drops of Valspar was then put on the clean surface, and then the plate glass disc, perfectly clean, was placed upon the drops of Valspar, care being taken that no air bubbles were included and that a sufficient amount of Valspar was used to insure a small excess oozing out at the edges. A weight of about two hundred grams was then placed upon the plate glass disc and small pieces of lead plate were used as anchors to keep the glass from sliding while the varnish was still wet. The covers remained untouched for a number of days, the temperature of the warming plate never being over 60 degrees centigrade. It should be added that the lids of the dishes were previously bored at the glass factory with a hole (B), large enough to admit the tip of a small pipette.

A thin layer of beeswax, paraffin or ozokorite (D) was put into the bottom of each dish, and to this an embryo or dissection (E) was fastened by means of glass needles. The dish was then filled with formalin solution, alcohol or other preserving fluid, so that the upper surface of the liquid (F) was somewhat higher than the lower surface of the plate glass disc (C). Lid (A) was now cemented on to the dish by museum jar cement (Sealo), and the opening (B) plugged with a small stopper or a bit of beeswax. If there should be any subsequent loss of liquid from the dish, the plug can be removed and a small mouthed pipette used to replace the liquid through this opening.

The advantages of such a dish are evident. The air space is entirely out of the field of view, and there is no condensation of moisture on the under surface of the part of the lid used under the microscope.

S. I. KORNHAUSER
DEPARTMENT OF ANATOMY,
UNIVERSITY OF LOUISVILLE

A STARCH TEST IN PHOTOSYNTHESIS EXPERIMENTS

A SEARCH for better methods of conducting routine experiments in photosynthesis has led to results that may prove useful to others. So far as I am aware these exact methods have not been reported before.

We find that with a number of leaves such as those of the horseshoe geranium the chlorophyll may be dissolved much quicker by first rinsing or lightly rubbing them in gasoline. Kerosene seems to be somewhat useful but is not as good. While we have not determined the reason we find that hot alcohol penetrates many times as rapidly after this treatment, thus very quickly and evenly extracting the chlorophyll. The extreme brittleness from the alcohol treatment may be overcome by immersing the leaves for a moment in water.

With this method we find that ordinary white vaseline is excellent for coating the leaves instead of the various waxes commonly used to prevent the entrance of carbon dioxide in certain experiments. The vaseline is quickly dissolved by the gasoline treatment, thus permitting rapid penetration of the alcohol and iodine solutions.

A *fresh* solution of iodine in gasoline gives a clearer starch test than does the iodine solution in potassium iodide or in alcohol. It is, however, subject to a slow deterioration and becomes inactive after a week or two. It should be mixed in small quantities as needed.

To facilitate the use of this method a schedule is given below:

(1) Wash in gasoline one minute.
(2) Clear in hot alcohol until leaves are white. If needed change alcohol as soon as green with dissolved chlorophyll.
(3) Water momentarily to relieve brittleness.

(4) Iodine solution. A small crystal of iodine in 30 cc to 100 cc of gasoline.

FRED W. EMERSON

PENN COLLEGE,
OSKALOOSA, IOWA

SPECIAL ARTICLES

THE ROLE OF KINOPLASM IN THE GENESIS OF VACUOLES

THE genesis of vacuoles is a subject which is receiving much attention at the present time. Nassanov, Ludford, Uhlenhuth and Bowen, for example, have studied the development of secretory vacuoles in *Protozoa* and in gland cells of higher animals, and a host of workers, as exemplified by recent letters to *Nature,* are busy with the origin during oogenesis of various kinds of yolks—all of which, since they lie in pockets in the cytoplasm, are probably definable as vacuoles or more strictly the content of vacuoles—while Guillermond and his school and recently Bowen have investigated the inception of the "vacuome" in plant cells. It is argued by many, though Bowen disagrees, that the contractile vacuole of the *Protozoa,* and also the plant vacuole, are homologous with the Golgi apparatus of metazoan cells; that fatty yolk at least as well as certain secretory vacuoles are a product of the same structure; but that other kinds of yolk and of secretory vacuole may arise from mitochondria and from nuclear extrusions. Whatever rôle these various structures may play in secretion of the vacuolar contents, our own observations on living cells, chiefly of plants, lead us to the conclusion that the actual formation of the vacuoles is the function of a more active labile and fundamental element of the cytoplasm of which one rarely hears mention at the present time, *viz.,* Strasburger's *kinoplasm* (active substance). Recent cytological research has concentrated on those bodies which give more or less characteristic staining reactions, but the above differentiation of the cytoplasm is also sufficiently characteristic in another way, *viz.,* in the behavior in life, to warrant, it would seem, the continued application of a distinctive title.

The kinoplasm is distinguished from the matrix by a slightly higher refractive index, by its origin and metamorphoses, and usually by its motility—characteristically, protoplasmic streaming and the translocation of other formed bodies in the cell is a function of the kinoplasm alone. The form most commonly assumed is filamentous, but Strasburger recognized the identity of the fibrillar with the film type of structure and his conception is supported by our own observations (1) of the origin of the fibrils from and (2) their retransformation into films or membranes. This relation, which bears on the subject of our title, is illustrated in more ways than one. Thus the kinoplasmic processes which lie within the cytoplasm, *e. g.,* in cells of *Spirogyra,* may frequently be observed to stream out from the demonstrably differentiated film of clear cytoplasm which envelopes the chloroplasts, or they may become attached to such a surface at both ends flowing out from one and into the other. Similarly, they may arise from the boundary of the nucleus where also a differentiated film of cytoplasmic substance can be shown to exist. The probable origin of other intracytoplasmic processes from the external or the vacuolar membrane is only inferred from what follows.

Similar in behavior and origin to the above are the little known pseudopodium-like processes which commonly fringe the central sap cavity in plant cells. Here again by extending across the vacuole the filaments may flow both into and out of a differentiated film, the vacuolar membrane or tonoplast. While there is, no doubt, some difference in character between films and filaments which are in contact with the cell sap and those which are internal to the protoplasm, their similarity of behavior seems to allow of the extension of the generic term kinoplasm to cover both. (Of course the more massive strands which cross the vacuole may contain several streams of kinoplasm with matrix between.) We may, with Strasburger, include also cilia and fine pseudopodial extensions of the plasmatic membrane on the exterior of naked cells, but to extend the term as he did to nuclear elements (hypothetical at that) is to destroy its usefulness.

All this, however, is somewhat aside from our main point, which is that the typical kinoplasm within the cytoplasm becomes transformed on occasion into vacuoles or at least into the lining membranes of such. For example, during conjugation in *Spirogyra* the gametes get rid of water by means of "contractile" vacuoles.[1] These first become conspicuous as mucilaginous looking globules which in turn are formed by condensation of the kinoplasm. A similar formation of vacuoles—which also may be contractile, *i. e.,* may burst—frequently takes place under more pathological conditions, as for example, when cells are acted on by a strong plasmolysing agent which penetrates somewhat, or by a narcotic. In such cases the kinoplasm may give rise to vacuoles by other mechanisms than preliminary sphere formation. The erstwhile streaming processes may fuse into a honeycomb structure, or the various membranes, the vacuolar and chloroplastic envelopes particularly, may develop blisters in their substance.

In suggesting that these observations on the origin of vacuoles be taken into account in the study of yolk

[1] Lloyd, Trans. Roy. Can. Inst. 15 (2) 1926.