the screen may be flooded with one color, with a shadow caster between the disc and this source, while a different color is used to illuminate the disc alone.

The advantages of this device over others previously tried by the writer are numerous. First of all, the students can see everything that is done; there are no concealed light sources, no elaborate equipment. An equally important feature is the fact that the colors seem to be in the object viewed and not merely cast upon the wall or diffused from behind through milk glass. There are no fringes of monochromatic light, which often serve to reduce the vividness of the mixture. It is true that such fringes may be removed by observation through a reduction screen, but such a procedure always tends to destroy the appearance of surface color to a considerable degree. A further advantage is the speed with which variations in the demonstrations may be set up; one of the most annoying things about color wheels is their inefficiency in this regard. Should it be desired to move the apparatus from room to room, the screen could easily be given a wooden frame so that it could be moved about and set on classroom desks. Portability of the light sources would depend upon the type available. Finally, this apparatus affords a negligible storage problem when not in use.

References

1. BRUNSWIK, E. *Experimentelle Psychologie.* Vienna: Julius Springer, 1935. P. 96.
2. KARDOS, L. *Z. Psychol.,* Ergänzungsb. 23, 1934.
3. WOODWORTH, R. S. *Experimental psychology.* New York: Holt, 1938. Pp. 611–614.

Recent Procedures for the Study of Three-dimensional Shapes of Resting Cells, and a New Method for the Shape Study of Cells in Division

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Since detailed studies of three-dimensional shapes of cells in tissues are relatively recent, standardized techniques have not as yet been adopted. The first investigations (*2*) utilized essentially Born's wax plate method. In this, individual cells were reconstructed from serial sections and examined under the compound microscope, the sections being modeled consecutively in sheets of wax. Subsequently a wax impregnation technique was employed (*5*), in which masses of cells were infiltrated with a hard wax. The wax casts of individual cells were then separated, studied under a binocular dissecting microscope, and modeled face by face.

In later work cells were analyzed in tissues, without sectioning or separation. In some cases the tissues were killed, stained either with safranin or Delafield's hematoxylin, cleared in xylol, and mounted in mineral oil. They were then studied under the binocular dissecting microscope. In another instance (*1*) living cells were stained lightly with neutral red, mounted in water, and studied similarly. All of these investigations have dealt with relatively large and mature cells.

Although the binocular dissecting microscope is well adapted to three-dimensional cell shape studies, the possibility of using it is eliminated in some of the most fundamental parts of the plant, such as the apical meristems, because of insufficient magnification. In the study of the three-dimensional shapes of minute cells, two different techniques have been used. In the first, the Duchartre-Lewis method (*3, 4*), longitudinal and transverse sections of the tissues were made; from these the number of faces, in section, could be determined under the compound microscope and the average number of faces per cell computed. While this method is helpful in determining the average number of faces, it has obvious limitations: it works best in tissues with the cells regularly arranged, it does not tell the precise number of faces on any individual cell, and it gives no information on the kinds of faces (whether they are quadrilateral, pentagonal, etc.). In the other investigation of minute cells (*6*), dealing with the apical meristem, stem tips were cleared in lactic acid (*8*), stained in Delafield's hematoxylin, and mounted "in toto" in balsam. They were then studied under the compound microscope.

Finally, in investigations now under way, the three-dimensional shapes of dividing cells in telophase have been analyzed. More specifically, the problem to be solved was determination of the original shape of the individual cells before division and of the two daughter cells directly after division, in order to make comparisons of the shape changes involved in the process of mitosis. To accomplish this it was necessary to see clearly the nuclei, both resting and dividing, the spindle fibers and cell plate, and the original walls of the cells. However, since the walls of several superimposed layers of cells had to be identifiable, merely by focusing, they could be stained only lightly. Furthermore, the dense cytoplasmic contents of the meristematic cells had to be nearly or quite transparent so that the components to be studied would not be obscured. To achieve the desired results the preparations were stained with Feulgen, ruthenium red, and orange G. The procedure was as follows:

Stem tips of *Anacharis densa* (Elodea) were dissected out, killed in Craf fixative, embedded, and sectioned at 25 μ or thicker. They were then stained with the Feulgen technique as outlined by de Tomasi (*9*), except that they were hydrolyzed longer (30 min) in N HCl and stained in decolorized fuchsin for 24 hrs. After being rinsed in distilled water the slides were put into a strong aqueous ruthenium red solution for 30 min, quickly rinsed again in distilled water, and run as rapidly as possible into absolute alcohol. They were then stained for about 1½ min in orange G in absolute alcohol and clove oil, rinsed in clove oil, put into xylol, and mounted in neutral balsam.

By this procedure the chromatic figures of the dividing nuclei were stained deep purple, while the color of the resting nuclei was much less intense. The middle lamella was darkened with ruthenium red, while the spindle fibers and cell plate were stained, to some extent with orange G.

The cellulose walls and protoplasm were not deeply colored, so that the shapes of several superimposed layers of cells could be determined effectively.

The slides were studied under oil immersion with a binocular monobjective Zeiss microscope, using Köhler illumination (7) and green and orange Wratten filters. The preparations are beautiful. The data obtained will be published elsewhere.

References

1. Dodd, J. D. *Amer. J. Bot.*, 1944, **31**, 120.
2. Lewis, F. T. *Proc. Amer. Acad. Arts Sci.*, 1923, **58**, 537.
3. Lewis, F. T. *Science*, 1928, **68**, 625.
4. Marvin, J. W. *Amer. J. Bot.*, 1939, **26**, 487.
5. Marvin, J. W., and Matzke, E. B. *Amer. J. Bot.*, 1939, **26**, 101.
6. Matzke, E. B. *Amer. J. Bot.*, 1948, **35**, 323.
7. Muñoz, F. J., and Charipper, H. A. *The microscope and its use.* New York: Chemical Publishing Company, 1943. P. 69.
8. Simpson, J. L. S. *Stain Technol.*, 1929, **4**, 131.
9. de Tomasi, J. A. *Stain Technol.*, 1936, **11**, 137.

An Improved Moist Chamber Slide for Use in Micromanipulation

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The moist chamber slide to be described here is essentially a modification of that described by Chambers and Kopac (*1*). Although the chamber considered here is designed for use with the Chambers micromanipulator, its basic construction features may be adapted readily to any chamber type according to individual preference or need (*1*).

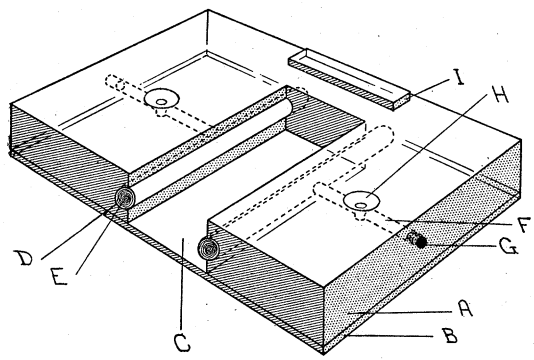

Fig. 1

The moist chamber slide consists primarily of a sheet of plastic (Fig. 1A), $2'' \times 3'' \times \frac{3}{8}''$ thick, cemented by means of Clarite to a standard 1-mm-thick $2'' \times 3''$ glass microscope slide (B). A centered, rectangular-shaped notch cut in the front edge of the plastic sheet constitutes the moist chamber proper (C). A rounded groove (Figs. 1D, 2) running the length of each side wall of the moist chamber holds a small roll of filter paper (E). Small drilled channels (F) extending from each edge of the plastic sheet and opening into their respective grooves in the side walls of the moist chamber (D) also hold small rolls of filter paper. The outside ends of these channels are closed by means of small stoppers (G). Each lateral channel (F) opens onto the top surface of the plastic sheet through a vertical funnel-shaped channel (Figs. 1H, 2). A small plastic strip welded on the top surface near the rear end of the moist chamber constitutes a coverslip stop (I).

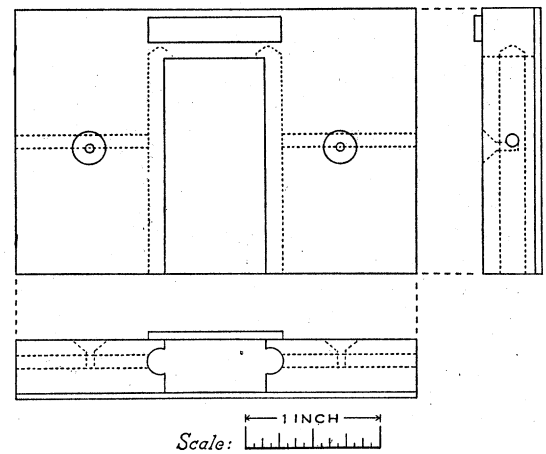

Fig. 2

In ordinary use, the chamber is moistened simply by the periodic addition of drops of water to the funnel-shaped channels. The filter paper rolls, by capillarity, quickly conduct the water directly to the critical area of the moist chamber. If one is studying simple qualitative influences of diverse water-soluble gases on the cells in the hanging drop, aqueous solutions of such gases also are introduced into the moist chamber in the same way, but the funnel-shaped channels are covered in between times by small coverslips sealed down with vaseline to prevent the diffusion of gas from these points. This method of keeping the chamber moist has several distinct advantages over the standard moist chamber slide. It precludes the chance disarrangement of the manipulation needles when in use. Further, it provides a measurable control of the relative amount of moisture within the chamber when this is essential. It dispenses with the interferences so commonly occasioned by filter paper strips within the chamber and excess water on its floor.

The detailed measurements necessary for the construction of this moist chamber slide may be obtained by applying the 1″ scale to the construction drawings (Fig. 2). The various properties of sheet plastic make it a material of choice and one very easily worked. Its uniform thickness virtually eliminates the necessary but usually laborious task of leveling the walls of the chamber.

Reference

1. Chambers, R., and Kopac, M. J. In *Microscopical technique.* C. E. McClung. (Ed.) New York: Paul B. Hoeber, 1937. Pp. 62–109.

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