The National Research Council will be ready to consider further requests for research assistance this fall. Applications should be filed with the Committee on Grants-in-Aid before October 15, 1933. Action upon these applications will be taken during the latter part of December.

ISAIAH BOWMAN,
*Chairman*

# SCIENTIFIC APPARATUS AND LABORATORY METHODS

## LAMELLIBRANCH LEUCOCYTES AS LIVING MATERIAL FOR CLASSROOM DEMONSTRATION

INCIDENTAL to studies concerned with the vitality of Molluscan leucocytes *in vitro* it was noted that those of *Ostrea virginica* possessed several features especially recommending them for classroom use as demonstration material. These features were found to be also evident in Venus and Anodonta and are probably common to most other lamellibranchs.

After removing one valve, a capillary pipette may be thrust into the heart and a quantity of blood withdrawn. A single specimen will yield more than enough material for a large class. The leucocytes in a drop, when placed on a slide, contract to a spherical shape and clump together. In from five to ten minutes they begin to thrust out pseudopods and move away from each other, and will then remain active for at least several hours if the drop is sealed. Since these forms occur in a practically pure culture, if properly collected, they are particularly suited to student purposes.

If a few drops of the blood are placed in a dish of the same water from which the animals came, the leucocytes will go on living for several days without further attention.[1] As the leucocytes collect on the bottom they are readily recovered by means of a pipette when needed. The advantageous features of such material are evident and include the ease with which they are obtained, the simplicity of handling, the abundance of cells and their relative hardiness. As demonstration material they show amoeboid movement, ingestion of food, and in fact all the usual features for which free-living amoebae are customarily employed to illustrate. If left in a sealed drop of sera over night they will be found to have again contracted to a spherical form. The addition of some fresh water will revivify them a second time, at least. It seems that their length of life *in vitro* is almost directly proportional to the amount of fluid in their container.

The operative technique mentioned is not imperative, as these mollusks, if placed in a water that is allowed to warm slightly, will emit quantities of leucocytes. This is the well-known diapedesis or "bleeding" of oysters under suffocation.[2] Thus a single animal can be used repeatedly if a suitable aquarium is handy to which it may be returned. Samples taken this way frequently show contamination. For example, oysters obtained from a New York market yielded an *Amoeba* of the *limax* type from the exterior of the shell and *Valkampfia calkinski* or *V. patuxent* from the intestinal tract.[3] The latter two can be cultured easily on ordinary nutrient agar plates, yielding abundant parasitic material. Thus, such a demonstration may have a further advantage, showing on a single slide, from a single source, leucocytes, free-living and parasitic amoebae all migrated or ejected from one animal with no operative effort. Without considerable familiarity it is almost impossible to distinguish the living leucocytes from the parasitic amoebae.

Since the three genera, Ostrea, Venus and Anodonta, as well as other similar ones, are wide-spread, little difficulty should be encountered in obtaining material from various localities. The animals for most part are fairly hardy and may be kept in a suitable aquarium or obtained from a market or other sources as needed.

C. M. BREDER, JR.
NEW YORK AQUARIUM
R. F. NIGRELLI
NEW YORK UNIVERSITY

## EXAMINATION OF THE REVERSE SIDE OF MICRO-MOUNTS

ONE of the greatest needs of the acarologist, and others who mount minute arthropods on microslides, is to see the other side of their specimen. To turn the slide over and look through the one to two millimeters of glass is hardly satisfactory because the objectives are corrected for cover-glass thickness. Obviously, this may be overcome by securing an objective corrected for a cover-glass thickness of one or more millimeters. To have such a lens made up in America is prohibitive. Besides, the American four-millimeter objectives have such small working distances as to strike the slide before coming into focus. Finally I discovered that W. Watson and Sons, London, stock a four-millimeter (six-inch) parachromatic objective with a long working distance and that this objective

[1] Orton (1924, Fisheries Invest. Series II, 6) reported that the leucocytes of *Ostrea edulis* will live from 3 to 4 days in a dish, while we have had those of *Ostrea virginica* live for 6 days under similar conditions.

[2] This effect has been discussed by Orton (1924) ref. cited and Yonge (1928), *Jour. Mar. Biol. Ass.*, 15. The slight provocation that will cause this response can hardly be considered suffocation in the ordinary sense and certainly must take place frequently in a state of nature.

[3] Hogue (1915), *Arch. f. Protist.*, 35 (1922), *Jour. Exp. Zool.*, 35.