used. The pieces should be thoroughly shellacked. Any parts of the geological pattern may be painted on a fresh cast with the air-brush. This is done by protecting the surrounding parts of the cast with the pieces of the stencil. After a single color has been applied to several casts, and has become dry, other colors may be blown on in succession until the whole geological pattern has been applied to the casts. Lakes and major streams may be blown on with the air-brush through a sheet lead stencil which has been drawn down to fit the topography. Complicated stream patterns and printing require hand work.

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A METHOD FOR CONCENTRATING AND FIX-ING FREE-LIVING PROTOZOA ON COVER GLASSES

In making slides of free-living protozoa the greatest difficulty usually encountered is the sticking of large numbers of organisms on the slide or cover glass, as the case may be. This difficulty is greatly overcome by the following method for fixing on the cover glass. By folding paper with a porousness comparable to that of mimeographing paper, make a box about 20 mm deep and of a size into which the cover glass closely fits. Smear the cover glass thinly with egg albumen and place it face up into the box. Stand the box on blotting paper and fill it with

fixative to a depth of 4 mm, or more if the cultures are poorly populated. Into the fixative pipette an equal, or less, amount of water containing the protozoa. The fluid seeps out and is absorbed by the blotting paper. Because the seepage is uniform and relatively slow, the organisms are left securely fastened and evenly distributed on the cover glass. When only a thin film of the fluid still envelops the organisms, remove the cover glass and drop it face down into a dish of the fixative. After fixation, follow the usual procedure for the fixing solution and stain selected. Reverse the above process, when fixing rhizopods, by putting them into the box, allowing them to become attached and then adding the fixative.

The various fixing solutions and stains give good results with the above method. Navaschin's solution is particularly good for sticking the organisms on the cover glass. Since the crystal violet-iodine staining method leaves the cytoplasm almost colorless, yet stains the nucleus well, it is excellent for studying nuclear divisions in total mounts of protozoa. With this stain and the above method for fixing, the nuclear behavior in ex-conjugants of *Paramecium caudatum* is easily followed; the chromosomes are especially sharply defined.

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THE BLANDY EXPERIMENTAL FARM AND THE MILLER SCHOOL OF BIOLOGY UNIVERSITY OF VIRGINIA

SPECIAL ARTICLES

THE INCIDENCE OF THE DISEASE-PRODUC-ING AMOEBA (ENDAMOEBA HISTO-LYTICA) IN 1060 COLLEGE FRESH-MEN AND ITS SIGNIFICANCE

During November, 1933, the newspapers of the country carried items concerning an "outbreak" of amoebic disease in Chicago, with reports of 100 or more cases and a number of deaths. These reports gave the impression that the disease-producing amoeba (*Endamoeba histolytica*) is extremely rare and mostly confined to the tropics.

While it is true that amoebic disease is more prevalent in the tropics and that the incidence of the parasite tends to become lower in the more temperate regions, nevertheless, it has been amply demonstrated that this amoeba is world-wide in its distribution and is not uncommon in the more temperate climates. In fact, the first case of amoebic dysentery to be reported in the literature, by Lösch in 1875, was found in a northern locality, St. Petersburg (now Leningrad), Russia. Since then the disease and the parasite have been extensively studied in many parts of the world.

Craig¹ has called attention to the probability that between 5 per cent. and 10 per cent. of the population of the United States harbor *Endamoeba histolytica*, while, in our more southern states, Faust,² Meleney³,⁴ and others have found a still higher incidence of infection. In order to determine the incidence of *E. histolytica* and other intestinal Protozoa in college students, the present authors added an examination of a single ordinary stool to the regular medical ex-

¹ C. F. Craig, "The Amoebiasis Problem," Jour. Am. Med. Assoc., xeviii: 1615-1620, 1932; "The Pathology of Amoebiasis in Carriers," Am. Jour. Trop. Med., xii: 285-299, 1932.

² E. C. Faust, "A Study of the Intestinal Protozoa of

² E. C. Faust, "A Study of the Intestinal Protozoa of a Representative Sampling of the Population of Wise County, S. W. Virginia," Am. Jour. Hyg., xi: 371-384, 1930; "The Incidence and Significance of Infestation with Endamoeba histolytica in New Orleans and the American Tropics," Am. Jour. Trop. Med., xi: 231-237, 1931.

³ H. E. Meleney, "Community Surveys for Endamoeba histolytica and Other Intestinal Protozoa in Tennessee," Jour. Parasitol., xvi: 146-153, 1930; H. E. Meleney, E. L. Bishop and W. S. Leathers, "Investigations of Endamoeba histolytica and Other Intestinal Protozoa in Tennessee." "III. A State-wide Survey of the Intestinal Protozoa of Man," Am. Jour. Hyg., xvi: 523-539, 1932.