TWO COLOR INTRAVITAM STAINING OF COLPODA DUODENARIA

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In *Colpoda duodenaria* Taylor and Furgason, methylene blue, C.I. 922, applied as Loeffler's methylene blue will color the contents of the food vacuoles dark blue in 10 to 30 minutes while little or no stain enters the cytoplasm. In contrast to methylene blue, a textile dye ("Rit", brand cardinal red, patent No. 2,042,473) will stain the cytoplasm of *C. duodenaria* a uniform pink in the same length of time. For this technique, "Rit" is prepared by stirring an excess with absolute methyl alcohol and decanting the saturated solution of stain from the excess dye and mineral salt present.

By simultaneous use of methylene blue and "Rit," the author has succeeded in double-staining living cells of *C. duodenaria*. For this purpose a methylene blue smear is made on a slide and a cardinal red ("Rit") smear on a cover glass. The smears are allowed to dry before use. When a large drop of culture containing *C. duodenaria* is placed on the smear on the slide and the cover glass is added, most of the organisms will have pink-colored cytoplasm and blue food vacuoles within ten to thirty minutes. The two color effect will not last more than an hour due to the entrance of methylene blue into the cytoplasm.

The technique used is similar to that described by Sabin (2) for human leucocytes. Although this method results in stain concentrations ten to a hundred times higher than usually recommended, the results appear to be quite satisfactory. The physiological interpretation of intravitam staining effects has been discussed by the author in a previous publication (1).

LITERATURE CITED

1. REETZ, C. E. 1948. Intravitam staining of Protozoa. Bull. A. S. A. M. 3:39.
2. SABIN, FLORENCE R., 1937, Supravital stains in McClung's handbook of microscopical technique. New York: Hoeber.

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