OCCURRENCE OF MOLDS AND YEASTS IN DAIRY PRODUCTS

by

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In literature on dairy and food microbiology, as represented by reference and text books by Tanner (1944), Hammer (1948), and Frazier (1958), the technique for separating bacteria from yeasts and molds in the examination of dairy products relies heavily on the acidification of media. This is reflected in the protocols adopted by the 11th edition of Standard Methods for the Examination of Dairy Products (Amer. Publ. Health. Ass. 1960), which also specifies acidification of media in which fungi of any types are to be examined.

As early as 1944 Smith & Dawson (1944) used rose bengal, a vital stain, for slowing down the rate of growth of fast-growing molds in mixed primary isolation media used in soil-population sampling. This dye also eliminated some types of bacteria. In 1950, Martin (1950) improved this medium by using penicillin and streptomycin, and in 1954 Cooke (1954, 1963) added Aureomycin* for substrates in which large numbers of bacteria could be expected.

* "Mention of commercial products does not imply endorsement by the Federal Water Pollution Control Administration, the National Center for Urban and Industrial Health, the U.S. Department of the Interior, or the U.S. Department of Health, Education, and Welfare."

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Because a medium containing rose bengal and antibiotics had yielded large numbers of colonies and species of fungi from soils, polluted waters, etc., in the Fungus Studies Laboratory, we decided to determine the utility of such a medium in the recovery of fungi from dairy products.

METHODS

Samples of cheese, butter, buttermilk, instant non-fat dry milk, sweetened condensed milk, whipping cream, homogenized milk, skim milk, half and half, chocolate dairy drink, and coffee cream were obtained by random over-the-counter purchase in grocery stores; samples of ice cream stabilizers and flavorings were obtained from laboratories of dairies producing ice creams, and raw milk and cream were obtained directly from dairies.

The samples were prepared and plated according to the techniques prescribed by Standard Methods (Amer. Publ. Health Ass. 1960), and according to the technique developed in the Fungus Studies Laboratory. Potato dextrose agar (Difco*), and Cooke Rose Bengal Agar (Difco*) were used in tandem, after rehydration according to the manufacturer's labeled instructions; an antibiotic was then added to the latter medium. Chlortetracycline hydrochloride (Aureomycin*) was obtained from the manufacturer, Lederle Laboratories, and one gram of the product was dissolved in 150 ml of distilled water. This was stored in the refrigerator and sterilized using a membrane filter prior to each use. The optimal concentration of Aureomycin in rose bengal agar, 35 µg/ml of medium, was obtained by either of two methods: (1) 0.5 ml of the stock solution was added to 10 ml of agar prior to pouring each plate, or (2) 1.0 ml of Aureomycin was added to 200 ml of medium after autoclaving. "Standard Methods" procedures require pH adjustment of potato dextrose agar to 3.5 ± 0.1 with 10 % tartaric acid; however, it is not necessary to adjust the pH of rose bengal agar (pH 6.0) because of the presence of rose bengal and Aureomycin. In some of the first studies, rose bengal agar plates were incubated at room temperature (22—30° C) for 7 days; later, for better control of incubation temperatures, plates were held in an incubator at 25° C (for the same incubation time). Potato dextrose agar plates were incubated at 20° C for 5 days. At the end of the incubation periods the colonies on plates of each test medium were counted and recorded, and those plates with yeasts or molds were transferred to the Fungus Studies Laboratory for identification of the fungi involved. Colonies appearing on both types of media were either sight-identified or transferred to slants of neopeptone-dextrose agar for later identification studies.

The mold-type fungi were identified by the usual morphological procedures, with special attention to spore and sporophore types. Techniques for identification of yeasts are more complicated because reliance is placed not only on morphological characters of the gross