Plate-Slide for the Culture and Morphological Observation of Fungi

MARIAN W. RICHTER AND DANIEL AMSTERDAM

Department of Microbiology, Kingsbrook Jewish Medical Center, and the Isaac Albert Research Institute, Brooklyn, New York 11203

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Adaptation of a polystyrene plate-slide is extremely well suited for the simultaneous macroscopic and microscopic diagnosis of fungi. This technique obviates the disorientation of identifying structures when cultures are mechanically disaggregated during preparation of slides for microscopy.

Laboratory identification of monomorphic molds still relies heavily upon the observation of in situ structures. A problem associated with fungal diagnosis has always been the need for a vessel which permits the concomitant growth of reproductive structures and yet allows for their examination by microscopy.

FIG. 1. A, Unfilled Petrislide. B, Petrislides filled with cornmeal agar and restored in their original packing container. C, Developing colony in the center of a Petrislide.

Heretofore, slide cultures (2) and specialized petri dishes (1, 3) have been proposed for this purpose. This report will describe a commercially available, modified petri dish which we have found best suited for the objectives outlined above.

The polystyrene plate-slide ("Petrislide," available from the Millipore Corp.) was originally developed for bacterial counting procedures. When filled with a selected medium (2.5-ml capacity), it is extremely well suited for the simultaneous macroscopic and microscopic diagnosis of the mold forms of fungi. The Petrislide is a unique combination, a slide (52 × 75 mm) upon which a petri dish (45 × 5 mm) is bonded. The dish portion of the unit is equipped with a tight-fitted closure which prevents evaporation and possible loss of spores, and the design is such that the entire unit can be placed on the microscope stage and readily viewed. Petrislides are not marketed as "ster-
ile”; however, we have never experienced any contamination.

In this laboratory, plate-slides are routinely filled with Sabouraud dextrose agar or cornmeal agar, although any medium suitable for fungal growth may be used. Medium can be poured into the bottom or the cover of the apparatus with equally advantageous results. When dry, plate-slides are inoculated in duplicate at the center of each dish so that one colony will be formed. After a suitable period of incubation, the entire growth chamber is placed on the microscope stage and is scanned with a ×10 objective. If greater magnification is required, the cover (or bottom) is removed, the growth is overlaid with lactophenol cotton blue stain, and a circular cover slip is placed over the entire area. Because lactophenol cotton blue stain will inhibit further growth, after viewing as described above, the unit should be discarded. However, the replicate plate will insure continued growth and development of reproductive structures. This technique eliminates the disorientation of identifying reproductive structures when cultures are mechanically disaggregated during preparation of slides for microscopy and affords simultaneous macroscopic and microscopic viewing of the fungus in its “natural” growth conditions.

As an alternative procedure to viewing with low power (×10) only, the mature colony can be examined directly without removing the cover of the Petrislide. In this procedure, no lactophenol cotton blue or cover slip need be added, and all indentifying structures remain undisturbed.

Figure 1A is a photograph of an unfilled, open Petrislide. The filled plate-slides can be stored in a horizontal position at 4°C in the original plastic packing container to conserve space (Fig. 1B).

An inoculated chamber containing a single large colony in the center of the dish is seen in Figure 1C. Figure 2 depicts the Petrislide in place underneath the objective lens on the microscope stage.

Although we have described a culture vessel which is well suited for the examination of filamentous fungi, the method is equally adaptable for the routine cultivation of unicellular yeast forms. However this method is not recommended for the cultivation of infective and easily aerosolized fungi such as Histoplasma capsulatum and Coccidioides immitis.

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