

Method for obtaining mycelial pads

C. R. Wrathall

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Abstract

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Wrathall, C. R. A method for obtaining mycelial pads of *Neurospora*.

Three types of culture conditions are generally used for the cultivation of *Neurospora* - submerged, shake and agar surface. Of the three, the surface growth is the most difficult to recover for analysis. The use of membroner, such as cellophane and Millipore filters can lead to questionable results, while scraping the agar surface is difficult and time-consuming. In the course of a current investigation of the sexual cycle of *Neurospora*, a technique for obtaining surface-grown mycelium has been used to great advantage.

A soft agar substrate, made by using Difco agar at a concentration of 0.2%. produces a medium with the consistency of thin "Jello". Growth will take place on the surface of this agar without penetration of the medium. A mycelial pad of sufficient strength to be manipulated is formed in about 60 hours on Synthetic Crossing Medium in a 100 mm diameter petri dish.

Harvesting can be accomplished in one of two ways. 1) The mycelial pad is separated from the sides of the petri dish in which it has been grown and water is run under the agar. The dish is then closed and inverted. The agar surface is then up and it can be loosened and washed away, leaving a circular pad which can be picked up on a piece of circular filter paper of the appropriate size. 2) The pad and agar are poured into a cheesecloth filter, washed with running water and squeezed to remove excess water. This wash is repeated two or three times. The resulting mycelia will be free from agar.

I have found the first of the two methods to be highly satisfactory for obtaining mycelial pads when analysis of portions of each pad is desired. ■ ■ ■

■ ■ ■ Biology Department, Rochester Institute of Technology, Rochester, New York 14623.