de Serres, F.J. A simple device for rapid preparation of conidial suspensions of Neurospora.*

Heterokaryon and other simple screening tests often require the preparation of a large number of conidial suspensions from different mutant strains of Neurospora. Our approach to this problem is to use the suspensor described below (a modification of a device designed originally by K. C. Atwood and T. H. Pittenger) which makes it possible to make large numbers of suspensions of conidia (1) uncontaminated by constituents of the culture medium, (2) rapidly, and (3) with the possibility of cross contamination reduced to a minimum.

The suspensor shown in the diagram is made out of 8-mm glass tubing, with an egg-shaped bulb blown about 6 inches from the top end. A hole 6-mm in diameter is placed 2-1/2 inches from the end for suction control with the forefinger. The open end of the inverted U-shaped tube (see diagram) inside the bulb should be placed near the base of the bulb so that a suspension of conidia can be completely removed. The tip is drawn out into a tube 2 mm in diameter (O.D.) about 6 inches in length and bent at an angle of 45° 1/2 inch from the end. The bent tip and inverted-U in the bulb should be in the same plane and perpendicular to the suction control opening at the top. The dimensions given are for a suspensor to harvest conidia from slants in 20 x 150 mm test tubes.

Suspensions are prepared in a hood. Sterile materials include two stainless steel containers filled with sterile distilled water, one ice-cold and the other boiling on a hot plate, test tube racks filled with (1) 13 x 100 mm tubes (with aluminum caps) each containing 2 ml water and (2) 20 x 150 mm tubes (with aluminum caps) containing 9 ml water; and a vacuum "safety" flask of at least 1-2 liter capacity.

To prepare a suspension, 2 ml of water is drawn up into the bulb, and the length of glass tubing from the tip into the bulb is dried somewhat by drawing air in slowly. The tip is then inserted into the culture tube and held over (but not in) the conidia, the suction being controlled and varied with the forefinger covering the hole near the top. With care, contamination by culture tube medium constituents can be avoided completely. The conidia should be drawn up the tubing and through the water in the base of the bulb. If the conidia come out above the water level, most will wind up in the waste water in the vacuum flask.

When all the conidia have been removed from the slant, the suspensor is disconnected from the vacuum tubing and the process of suspending these conidia is completed by drawing up additional water from a 20 x 150 mm tube. The suspension is blown out into this test tube and all of the conidia are removed from the device with repeated flushing. The suspensor is reconnected to the vacuum tubing and the entire device cleaned and sterilized between cultures by filling it up with boiling water and turning it up on end to drain the water out through the top. This process is repeated once or twice to rinse out all mycelial fragments and conidia and to inactivate any remaining. The device is then filled with the sterile cold water and emptied to cool it thoroughly before preparing the next suspension.

With this device a conidial suspension containing a total of 1-3 x 10^8 conidia can be prepared from a single 5-7-day-old culture grown on an agar slant in a 20 x 150 mm tube.

Figure 1. Device for rapidly preparing conidial suspensions.

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