

## RESEARCH NOTE

### Mass Culture of Mycophagous Nematodes

A. A. F. EVANS<sup>1</sup>

Biochemical and physiological studies on plant parasitic and mycophagous nematodes are often limited by insufficient nematode tissue. Although hundreds of thousands of nematodes may be obtained from monoxenic cultures of nematodes and host fungus on agar substrate (1), the total fresh weight of nematodes obtained, even from many such cultures, is usually small.

A method was developed for economically obtaining greater quantities of nematode tissue than has been produced by conventional methods. Forty grams of clean wheat seed is autoclaved with 200 ml tap water for 20 min at 15 psi in a wide mouth preserving jar cov-

ered with petri dish lids. After cooling, the wheat is inoculated with a monoxenic culture of nematodes on an appropriate host-fungus and then incubated at a suitable temperature until the nematode population has increased. Heat produced in the culture often necessitates setting the incubator at 1–5 C below the optimum for nematode population increase. Maximum weight of nematode tissue is reached when the wheat substrate darkens and nematodes leave the culture to aggregate on the walls of the container (Fig. 1). For each combination of fungus and nematode, experimentation with inoculum size and timing is necessary for maximum yields. Too many nematodes in the inoculum may kill a slow-growing fungus; in such cases it is usually better to establish the fungus alone first and add nematodes later. The cultures are readily harvested by first

Received for publication 19 June 1969.

<sup>1</sup>Waite Agricultural Research Institute, Glen Osmond, South Australia 5064. Present Address: Department of Nematology, University of California, Riverside, California 92502.

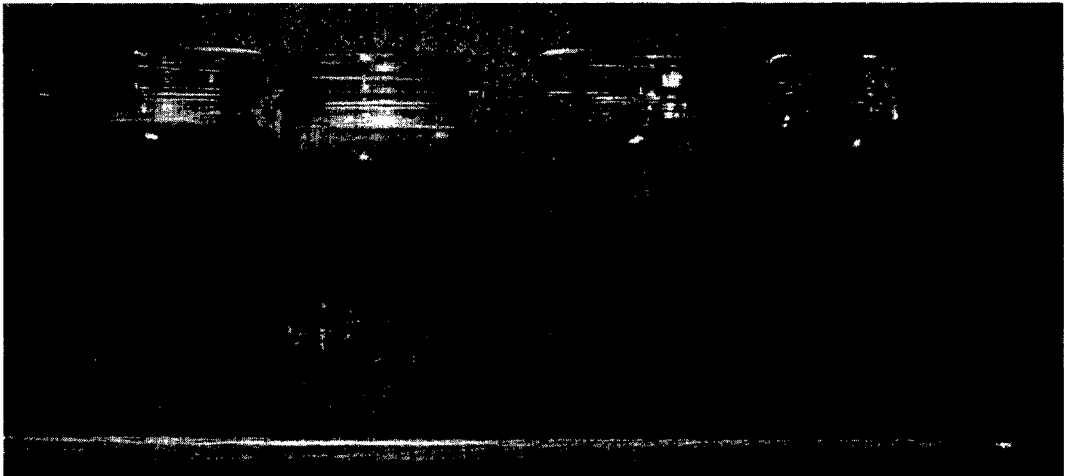


FIG. 1. Cultures of *Aphelenchus avenae* using *Rhizoctonia solani* as host-fungus. The two jars on left contain young cultures; two jars on right show the exhausted wheat-fungus substrate with nematodes aggregating high on the walls of the jars.

washing nematodes from the inner walls of the jar and then soaking the matted wheat-fungus residue in water to liberate remaining nematodes.

Using this method, 6 jars of *Aphelenchus avenae* feeding on *Rhizoctonia solani* at 25 C yielded an average fresh weight of 1.22 g of nematodes per jar (average dry weight, 0.39 g) after an incubation period of 6 weeks (Fig. 1). *Ditylenchus myceliophagus* has been grown on *Agaricus bisporus* and *D. destructor*, *Aphelenchoides saprophilus* and *A. bicaudatus* have been grown at 5–30 C on an unidentified fungus isolated from soil. The average size of nematodes of most species grown by this method has been greater than those grown on the same fungus in agar cultures.

Contamination by bacteria or yeast is the most common cause of low yields; great care should be taken to ensure that only clean cultures are used as inoculum. Attempts to increase the amount of wheat substrate used, and therefore the yield of nematodes from each jar, have met with little success. It may be that diffusion of heat and carbon dioxide from the culture is restricted if the volume becomes too large, although the possibility remains that further modification of the technique will result in larger yields of nematodes.

#### LITERATURE CITED

1. GOODEY, J. B. 1963. Laboratory methods for work with plant and soil nematodes. Great Britain Ministry of Agriculture, Fisheries and Food. Tech. Bull. 2, 72 p. Her Majesty's Stationery Office, London.