SIMPLE SCREENING METHODS FOR ASSESSING THE PREDACIOUS ACTIVITY OF NEMATODE-TRAPPING FUNGI

BY

S. GALPER, L. M. EDEN, G. R. STIRLING and L. J. SMITH
Queensland Department of Primary Industries, Meiers Road, Indooroopilly, Queensland 4068, Australia

Australian isolates of nematode-trapping fungi were screened for activity against nematodes on agar and in soil. All isolates produced traps extensively on agar in response to Caenorhabditis elegans and, within 1-3 days, populations of this nematode were generally reduced by at least 90%. Juveniles of Meloidogyne javanica induced fewer traps on agar and most fungi took more than 3 days to reduce numbers of nematodes by more than 90%. Experiments in microcosms showed that predacious activity on agar gave little indication of the capacity of a fungus to trap nematodes when added to mineral soil. Species such as Dactylella candida and Arthrobotrys dactyloides, which formed detachable rings and knobs or constricting rings, consistently reduced the number of M. javanica juveniles recovered from soil whereas network-forming species sometimes did not. In studies using agar 'sandwich' and buried slide techniques to monitor trapping activity in soil, D. candida and A. dactyloides consistently produced traps within 5 days of being introduced, whereas network-forming species produced few traps in soil and their trapping activity varied from experiment to experiment.

Keywords: nematode-trapping fungi, Dactylella candida, Arthrobotrys dactyloides, screening methods

Ever since Linford's pioneering work with the nematode-trapping fungi more than 50 years ago (Linford, 1937; Linford et al., 1938; Linford & Yap, 1939), there has been interest in using these fungi for biological control. Numerous experiments have been done with the nematode-trapping fungi but the results have been inconsistent (Stirling, 1991) and there are not yet any examples where a preparation containing these fungi is widely used for biological control purposes.

There are many reasons for the lack of success with the nematode-trapping fungi, but the process used to select isolates with biological control potential has contributed. Most isolates used in biocontrol studies appear to have been chosen at random from the diverse range of species in soil and there have been few attempts to compare the performance of isolates. Since the literature makes it clear that there is considerable diversity within the nematode-trapping fungi, particularly in terms of their trapping mechanisms, competitive saprophytic ability and spontaneity of trap formation (Cooke, 1963 a,b,c, 1964; Jansson & Nordbring-Hertz, 1979), it is therefore important that screening procedures are available to identify useful isolates.

A number of established techniques have potential for use in screening programmes with the nematode-trapping fungi but unfortunately none is entirely satisfactory. Determining trapping activity on agar (Heintz, 1978) is
simple and inexpensive, but such systems provide the fungus with a ready supply of nutrients and little competition. Also, free-living rather than plant-parasitic nematodes are usually used as a target and the agar provides an ideal surface for traps to form and for nematodes to come into contact with them.

Assessment of predacity in soil is likely to provide a more realistic indication of biological control potential. However, systems involving sterile soil (Hayes & Blackburn, 1966; Jansson, 1982) do not have the complexity of natural soil. An agar disc technique such as that developed by Cooke (1961) has some advantages, but it is sometimes difficult to quantify trapping activity because soil particles become embedded in the agar. In this work, we used three simple methods to assess trapping activity in soil and compared the results with those obtained on agar.

MATERIALS AND METHODS

Isolation and identification of nematophagous fungi
Soil was collected from private gardens and agricultural fields in Queensland, Australia. Soils where populations of plant-parasitic nematodes were relatively small and soils with a history of organic manuring were specifically targeted. Nematophagous fungi were isolated by sprinkling 1 g soil onto water agar (WA) and baiting the plate with 3000 Caenorhabditis elegans (Maupas) Dougherty, or by differential centrifugation (Barron, 1977). Isolations were also made from soils that had been enriched with nematodes by adding 10,000 C. elegans or 5,000 Meloidogyne javanica (Treub) Chitwood juveniles to 150 ml soil. After incubating nematodes for 5 days, they were recovered using a Baermann funnel and transferred to water agar that had been baited with 3,000 C. elegans. All plates were examined for trapped and inactive nematodes over a period of 5 weeks and nematophagous fungi were recovered by transferring spores to corn-meal agar (CMA). Isolates were identified by Dr. D. W. Minter, International Mycological Institute (IMI), Kew, U.K.

Growth in culture
Four replicate 9 cm diam Petri dishes containing 10 ml CMA were inoculated with a 4 mm diam disc of one of 18 fungal isolates, taken from the edge of actively growing colonies, and incubated at 27°C. Once the fungi had grown onto the agar, the dish was marked at the advancing edge of the colony. This process was repeated 2-7 days later, depending on the rate of growth of each fungus. Radial growth rates (mm/day) were then calculated.

The growth of isolates in liquid culture was compared by inoculating two replicate 250 ml Erlenmeyer flasks containing 100 ml GPY (15 g glucose, 2 g peptone, 5 g yeast, 1 g asparagine, 0.5 g K₂HPO₄, 0.25 g MgSO₄·7H₂O, 0.001 g thiamine HCl, 1 litre H₂O) with two 4 mm diam discs of each fungus cut from colonies on CMA. Flasks were incubated on an orbital shaker at 27°C...