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Scanning electron microscopy of Meloidogyne incognita juveniles entrapped in maize roots by a nematodetrapping fungus Arthrobotrys dactyloides

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During a study on the presence and location of *Meloidogyne* incognita race 2 within maize roots, a nematode-trapping fungus, *Arthrobotrys* dactyloides was observed for the first time on maize in South Africa.

Tydens 'n ondersoek na die voorkoms en posisie van Meloidogyne incognita ras 2 in mieliewortels, is 'n aalwurmvangende fungus Arthrobotrys dactyloides vir die eerste keer in Suid Afrika op mielies waargeneem.

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Fungi, including predacious fungi, are common in soil and are therefore found in close proximity to roots of agricultural crops. This is also the case with nematodes. In some instances the production of trapping devices by fungi is initiated by the presence of nematodes (Barron 1977). During a study on the presence and location of *Meloidogyne incognita* Chitwood race 2 within the maize root system, a

nematode-trapping fungus, Arthrobotrys dactyloides Drechsler, was observed for the first time on maize in South Africa. Meloidogyne species are known plant-parasitic nematodes on maize (Walters 1976; 1978; Kinloch 1983; Keetch & Buckley 1984; Storey 1985; Windham & Williams 1987; 1988). More than 50 species of nematode-trapping fungi have already been described (Balan & Gerber 1972). These predacious fungi constitute the order Zoopagales. The family Moniliaceae of the Hyphomycetes also contains many species of predacious fungi (Moore-Landecker 1982).

The *M. incognita*-infected maize roots were prepared for SEM according to the following procedure. Pieces of infected roots were fixed in Todd's fixative (Aldrich & Todd 1986) for six hours and washed in cacodylate buffer three times for 10 minutes each. The samples were post-fixed in 1% osmium tetroxide for one hour, washed for 10 minutes in distilled water and dehydrated in an acetone series (50%; 70%; 90%; 100%; 100%) for 15 minutes each. After dehydration, the roots were critical-point dried in liquid carbon dioxide, mounted on SEM-stubs, coated with gold-palladium and viewed in a Cambridge Stereoscan 250 SEM.

From results obtained in the SEM study, it is clear that the nematode-trapping fungus, A. dactyloides, entrapped juveniles in areas of the roots damaged by the feeding activity of Meloidogyne juveniles and females (Figure 1). Most of the damage occurred in the vicinity of secondary root bases.

Arthrobotrys dactyloides forms ring-like traps, consisting of three cells, which constrict to mechanically catch nematodes (Drechsler 1937). Death is caused by the closure of the trapping ring (Figure 2) as well as by ammonia produced by the fungus (Balan & Gerber 1972; Webster 1980). The trapping of free-living nematodes such is Rhabditis, Cephalobus and Acrobeles (Drechsler 1937) and

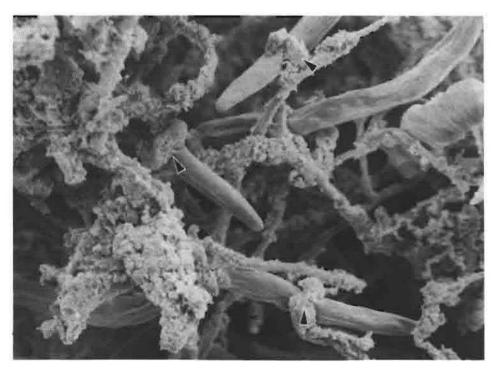


Figure 1 M. incognita larvae on maize roots trapped by A. dactyloides (arrows).

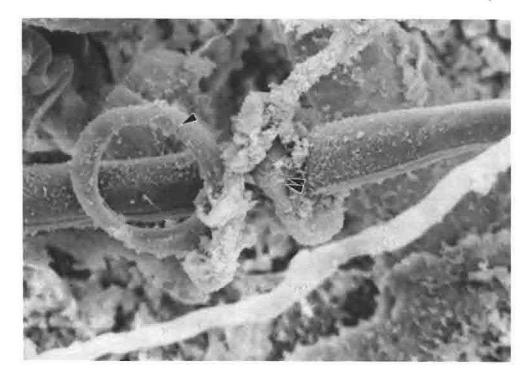


Figure 2 Deflated rings (single arrow) and inflated rings (double arrow) of A. dactyloides.

Panagrellus redivivus (Balan & Gerber 1972), and plantparasitic nematodes such as *Meloidogyne* spp. (Mankau 1980) has been described.

Agricultural soils normally have high populations of nematode-trapping fungi which only trap nematodes under unfavourable conditions (Barron 1977). The inducement of the ring-like traps is initiated mostly in response to the secretion of nemin compounds by the nematodes (Barron 1977). The trapping is achieved by the enlargement of the ring cells (Figure 3). One of the cells enlarges a fraction of a second prior to the other two cells (Webster 1980). This enlargement is accompanied by vacuolation of the cell contents and by elastic stretching of the inner wall of the ring, while the outer wall of the ring does not change shape (Estey & Tzean 1976). The ring closure is normally completed within one tenth of a second (Webster 1980).



Figure 3 The three enlarged cells (arrows) of the trapping ring.

Studies have been done to determine the feasibility of using predacious fungi for biological control of plant-parasitic nematodes. The addition of fungal spores alone to the soil, however, does not increase the population of predacious fungi that occurs naturally in soil (Moore-Landecker 1982). Furthermore, in our observations no ring-like traps were found inside the maize roots, only on the root surface and in the vicinity of the secondary root bases. This places limitations on the use of this fungus as a biological control method, as the juveniles are not trapped after penetration of the root system.

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Selective predation on *Protea neriifolia* (Proteaceae) seeds by small mammals

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The ability of small mammals to select *Protea neriifolia* seeds containing embryos (full seeds) was examined. Full and empty seeds were glued to wooden boards and placed at three sites in young fynbos. Small mammals were able to select and remove significantly more full seeds than expected. Seed removal rates varied between the study sites but this was not related to the density of small mammals trapped at those sites. Selective feeding greatly increases the feeding efficiency of small mammals because 90% or more of the seed dispersed after fires may be empty.

Die vermoë van klein soogdiere om *Protea neriifolia* sade met vrugkieme (vol sade) te selekteer is ondersoek. Vol en leë sade is op hout plankies vasgegom en op drie studieterreine in jong fynbos geplaas. Die klein soogdiere het betekenisvol meer vol sade verwyder as wat verwag was. Die tempo van saad verwydering het verskil tussen die studieterreine maar daar was geen verband met die digtheid van klein soogdiere wat gevang was nie. Deur selektief te voed kan klein soogdiere hul voedingsdoeltreffendheid aansienlik verhoog aangesien 90% of meer van die sade wat na 'n brand versprei is leeg kan wees.

Seed set in fynbos Proteaceae is low, generally less than 10% (Collins & Rebelo 1987). Protea neriifolia R.Br. has 270-320 florets per inflorescence (Collins & Rebelo 1987), but seed production is low, typically less than five per inflorescence in Swartboskloof. Many inflorescences contain no viable achenes, hereafter termed seeds, at all (Le Maitre unpub.). Many florets produce empty, sterile seeds which outwardly resemble embryo-filled seeds. However, seeds with a healthy white embryo (hereafter termed full seeds) are firm, but seeds with decayed embryos or without embryos (hereafter termed empty seeds) are easily compressed. Empty seeds may also have a dull appearance compared with the glossy look of full seeds. Ninety-five per cent or more of full seeds selected by the human hand on the basis of feel and appearance will germinate under controlled conditions (Le Maitre 1990). If small mammals can use similar criteria to distinguish full from empty seeds, without first opening them, their efficiency as seed predators would be significantly higher than if they fed non-selectively.

Seed predation by small mammals can have a significant impact on the regeneration of fynbos Proteaceae with canopy-stored seeds, both between and after fires (Bond 1984; Bond, Vlok & Viviers 1984; Breytenbach 1984; Bond & Breytenbach 1985). Most of these studies used small mammal enclosures in field experiments or were done with captive animals. In this paper we report on a field experiment using seeds of *P. neriifolia* and designed to test:

- (a) the ability of small mammals to select seeds;
- (b) whether removal rates differ between forest, ecotonal and burnt fynbos communities; and
- (c) whether the removal rate is related to the abundance of small mammals.

The study was carried out in the Swartboskloof catchment in the Jonkershoek Valley near Stellenbosch, in the southwestern Cape. The catchment was burnt on 17 and 18 March 1987. The pre-fire vegetation was a tall (>2 m), 29-year-old shrubland dominated by *P. neriifolia*. This study was carried out concurrently with other experiments which examined the impact of seed and seedling predation on seedling recruitment of *P. neriifolia* after the fire (Botha & Le Maitre 1992).

Seeds of *P. neriifolia* were collected before the fire and sorted by hand. The sorted seeds were stuck onto 120 × 120-mm masonite boards using wood glue. Four full seeds were stuck onto each board. Six empty seeds were glued in a ring around each full seed on half of the boards giving 0,17 full seeds per empty seed on boards with both kinds.

Ten boards, five with each type of seed arrangement, were placed alternately, about 5 m apart, along each of seven lines on 8 April 1987. The lines were spaced about 20 m apart. The first line was situated in riverine forest, the second in the partly burnt ecotonal community (about 2 m from the forest edge) and the remainder in burnt fynbos. A further thirty boards, fifteen of each type, were set out in a similar fashion at two burnt fynbos sites on 11 April 1987. The boards were visited at intervals of one to two weeks and the number and type, full or empty, of any missing seeds were recorded. The experiment continued until 6 July 1987 when it was found that heavy rains had soaked the boards, with the result that seeds had become unstuck or had germinated on the boards.

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