

## Diversity and trapping efficiency of nematophagous fungi from Oman

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**Summary.** A survey of the nematophagous mycobiota biodiversity of 82 soil and leaf-litter samples in the Sultanate of Oman yielded ten species of nematode trapping fungi belonging to three genera. The species are: *Arthrobotrys eudermata*, *A. thaumasia*, *A. musiformis*, *A. oligospora*, *A. oligospora* var. *oligospora*, *A. oudemansii*, *A. multiformis*, *A. javanica*, *Drechslerella brochopaga* and *Gamsylella geophyropaga*. This is the first record of these species in Oman. *Arthrobotrys multiformis* represents the second record of this species worldwide. A systematic study showed that *A. oudemansii*, *A. multiformis* and *A. javanica* were morphologically more variable than was so far known. In four days, *A. oligospora*, *A. thaumasia*, *D. brochopaga* and *A. eudermata* trapped all nematodes added to the Petri dishes (*Panagrellus redivivus*, 100 specimens per dish). No significant differences were found in the trapping efficiency among the species tested.

**Key words:** mycobiota, nematophagous fungi, nematodes, Oman, trapping effectiveness.

### Introduction

Nematophagous fungi are fungi that attack and feed on living nematodes or their eggs. The taxonomy of nematophagous fungi is under continuous revision and re-evaluation. The most important contributions to the systematics of this group have been made by Drechsler (1937), Subramanian (1963), Cooke and Dickinson (1965), Rifai and Cooke (1966), Haard (1968), McCulloch (1977), Schenck *et al.* (1977), Oorschot (1985), Barron (1989), Zhang *et al.* (1994), Rubner (1996), Li *et al.* (1998), and Zare *et al.* (2000). The most re-

cent revision was by Scholler *et al.* (1999) who proposed a new genus concept on the basis of biochemical, morphological and molecular features (rDNA sequence data).

The geography of the Sultanate of Oman is characterized by a variety of plains and mountains that are typical of the harsh conditions of arid and desert habitats. In such regions, nematophagous fungi have been poorly studied (El Amin, 1980; Muhsin and Kasim, 1998; Elshafie *et al.*, 2003, 2005). We carried out a survey in order to provide a better understanding of the morphological diversity characterizing the species inhabiting arid habitats and temperate and tropical areas.

Plant-parasitic nematodes occurring in the Sultanate of Oman parasitize various types of vegetable and fruit crops. Moghal *et al.* (1993) identified 35 parasitic nematode species attacking 35 plant

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hosts, causing mean annual crop losses in citrus production of 15%.

Fungi are one of the most important micro-organisms that can be used to control nematodes. About 70 genera and 160 species of fungi have been found associated with nematodes, but few of them are suitable for biological control because of their lack of effectiveness (Siddiqui and Mahmood, 1996). Few papers report have studied the effectiveness with which nematophagous fungi trap nematodes in Petri dishes or in the soil. Nematophagous fungi can live saprophytically as well as preying on soil nematodes with a variety of trapping organs. As predators, they form trapping structures with which they catch nematodes to exploit the nematode carbon and nitrogen contents and other nu-

trients they provide. Scholler and Rubner (1994) studied the predacious activity of *Arthrobotrys oligospora* in relation to the carbon (C) and nitrogen (N) concentration of the nutrient medium. They found that at certain concentrations of C and N, the fungus did not form traps and lived saprophytically.

The objectives of this study were to survey nematophagous fungi in Oman and compare their predacious activity, in order to identify fungi with a greater trapping effectiveness that could be used to control crop nematodes.

## Materials and methods

Soil samples were randomly collected from the rhizosphere of plants infected with nematodes. The sprinkled-plate technique described by Rubner (1996) was used to isolate nematophagous fungi, sprinkling one gram of soil each onto four Petri dishes containing water agar (WA). One hundred nematodes (*Panagarellus redivivus*, Linne Goodey) were added to the cultures and incubated for 1–2 months at room temperature (23°C). Trapped nematodes were then transferred to corn meal agar (CMA) consisting of 8.5 g CMA (Oxoid, Unipath Ltd, Basingstoke, England), 12.5 g additional agar (Oxoid) and 1 liter of water. Using a sterile human hair fixed onto a fine needle, single spores of nematophagous fungi were collected from the erect conidiophores, cultured on half-strength CMA and challenged with nematodes. The nematicide effectiveness of *A. eudermata*, *A. thaumasia*, *A. oligospora* and *Dactylaria brochopaga* was tested by adding about 100 nematodes to each Petri dish containing a five-day-old CMA culture of fungi, in triplicate plates. Trapped nematodes were counted with the aid of a stereoscopic binocular microscope over a period of four days. The trapping effectiveness of each species was compared using one way analysis of variance (SPSS ver. 10).

## Results and discussion

Although Oman has one of the world's most forbidding environments, some nematophagous fungi were found associated with different plants in various locations. Table 1 shows the location, source, incidence and trapping devices of ten nematophagous fungal isolates taken from 82 soil

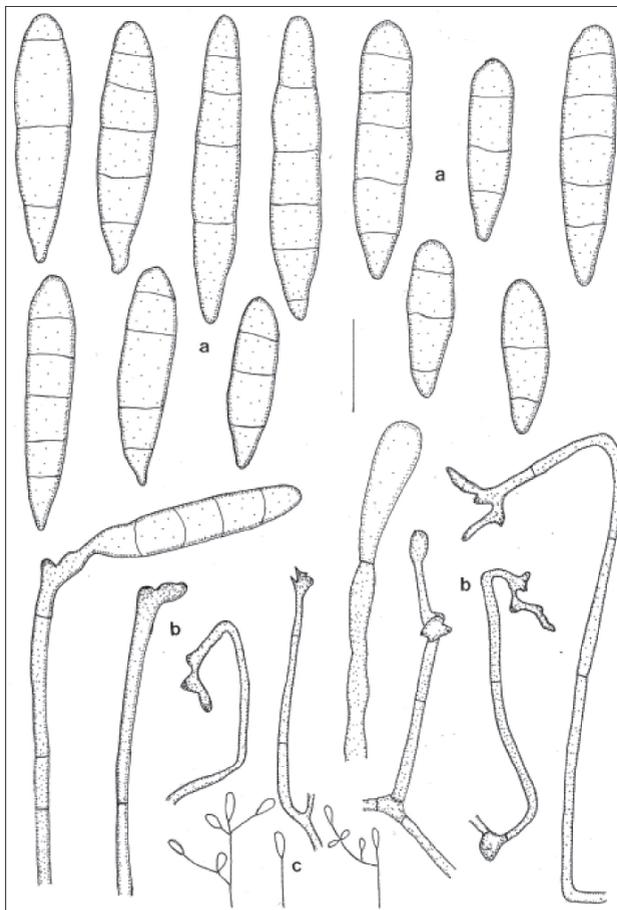


Fig. 1. *Arthrobotrys multiformis*: a. macroconidia; b, c. primary conidiophores. Habit sketch of conidiophore. Scale bar: a, b=25  $\mu$ m.

Table 1. Locations, source, incidence and trapping devices of nematophagus fungal species in Oman.

Trapping device	Incidence in soil (%)	Plant associated	Isolation location	Fungal species
3 dimensional adhesive networks	36.6	Compost, <i>Musa acuminata</i> , <i>Magnifera indica</i>	Nizwa, Barka, Al-Khoud	<i>Arthrobotrys oligospora</i>
3 dimensional adhesive networks	24.4	<i>Magnifera indica</i> , <i>Citrus limettioides</i> , <i>Panica granatium</i> , <i>Musa acuminata</i> , <i>Citrus aurantifoila</i>	Tiwi, Izki, Samail, Barka, Jabal Al-Akhdar and Rumais	<i>A. oligospora</i> var. <i>oligospora</i>
3 dimensional adhesive networks	14.6	<i>Magnifera indica</i> , <i>Phoenix dactylifera</i> , <i>Citrus aurantifoila</i> , <i>Musa acuminata</i>	Al-Khoud, Salalah, Rustag	<i>A. thaumasia</i>
1 to 3 cells adhesive columns	4.9	<i>Citrus aurantifolia</i>	Nizwa	<i>Gamsylella geophyropaga</i>
2–3 dimensional adhesive networks	3.7	<i>Ficus benegalensis</i>	Al-Khoud, Tiwi	<i>A. multiformis</i>
3 dimensional adhesive networks	3.7	<i>Musa acuminata</i>	Saham	<i>A. oudemansii</i>
3 dimensional adhesive networks	2.4	<i>Citrus aurantifolia</i>	Nizwa	<i>A. javanica</i>
2–3 dimensional adhesive networks	2.4	<i>Solanum melongena</i>	Tiwi and Izki	<i>A. musiformis</i>
3 dimensional adhesive networks	1.2	<i>Allium cepa</i>	Izki	<i>A. eudermata</i>
Stalked, 3 cells constricting rings	1.2	<i>Hibiscus esculentus</i>	Al-Rustag	<i>Drechlerella brochopaga</i>

and litter samples, obtained from different sites in Oman. The cosmopolitan nematophagous fungi *A. oligospora* and *A. oligospora* var. *oligospora* were the most frequently isolated, being recovered from 36.6 and 24.4% of soil samples respectively. Other species were rare, occurring in 1.2% to 15% of samples.

When they were examined microscopically, *A. oudemansii*, *A. multiformis* and *A. javanica* had a more varied morphology than has been reported in the literature so far (Fig. 1). Moreover, *A. oudemansii* (CBS 109510) had shorter and wider conidia, lacking microconidiophores and microconidia, when compared with the type species and all other described isolates.

*Arthrobotrys multiformis* (Sultan Qaboos University Herbarium, No. 44) was also rare. This is only the second record for this species world-wide (it was first described by Dowsett *et al.*, 1984, and re-examined by Rubner, 1996). It differs from the described species in having larger conidia (41–

145×8–16 µm vs. 39–90×4–7.5 µm), fewer septa (1–6 vs. 4–12) and longer secondary conidia (10–39×2–8 µm vs. 20–25×5 µm).

The conidia of *A. javanica* (CBS 109508) isolated in this study were slightly longer and narrower than the described isolates and not constricted at the septa.

*Arthrobotrys oligospora*, *A. thaumasia*, *D. brochopaga* and *A. eudermata* trapped all nematodes in four days (Fig. 2). Although the trapping effectiveness of these fungi varied in the first two days, the significant differences between them disappeared by day four ( $F=1.982$ ,  $P\pm 0.05$ ). In general, nematophagous fungi vary in their nematode trapping effectiveness. Gonzales *et al.* (1998) reported that in seven days *A. robusta* trapped 32.3% of added nematode larvae, and *Gamsylella geophyropaga* 93%. In Petri dishes containing solid culture *A. oligospora* trapped 90% of nematodes in 16–40 hours, while *M. thaumasia* required 40 hours to trap the same percentage of nematodes

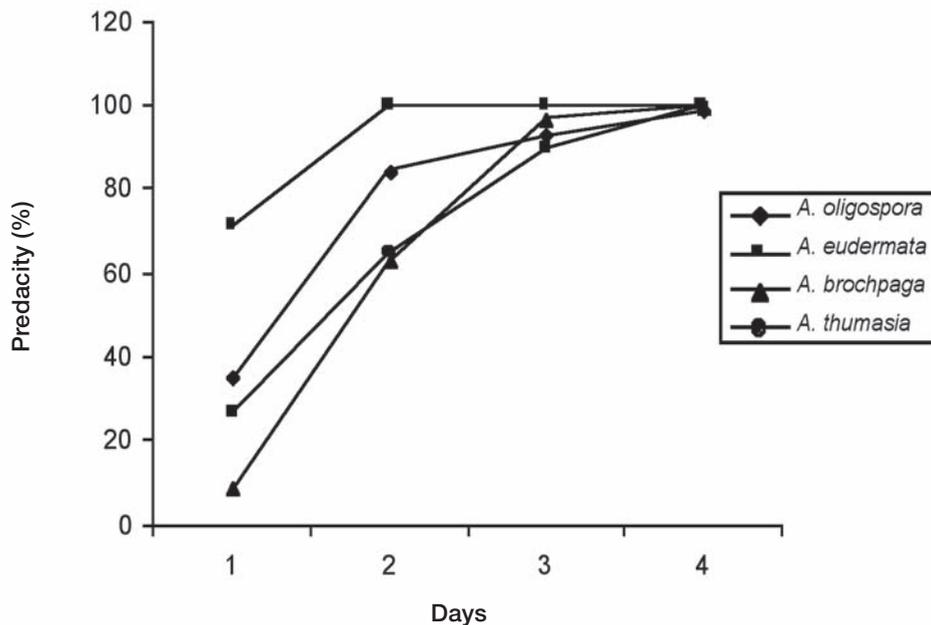


Fig 2. Comparison of trapping effectiveness (%) of four species of *Arthrobotrys* nematophagous fungi on specimens of *Panagrellus redivivus* added to each Petri dish.

(Rajeswari and Sivakumar, 1999). *A. oligospora* formed traps in liquid culture capturing 90% of nematodes (Friman, 1996; Rajeswari and Sivakumar, 1999).

The trapping experiment was carried out *in vitro* and the activity of fungi and nematodes in the soil may be different. In Petri dishes, the fungus is likely to capture all the nematodes added to the culture, but in the soil the nematodes have many opportunities to escape capture and remain alive. Furthermore, a truly effective antagonist should not eliminate its own source of nutrients (Jansson, 1982). Other biotic and abiotic factors also affect fungal activity and trapping effectiveness in the soil. These factors, alone or in combination, may explain the low success rate so far obtained in studies on nematode-trapping fungi. However, species and isolates adapted to the harsh environmental conditions that are normal in Oman, involving aridity, high temperature and high salinity may represent potential biological agents to control crop nematodes, which are wide-

spread in the country. Further data and tests are, however, required to investigate the activity of these fungi *in vivo* and their effectiveness in controlling nematode populations.

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Accepted for publication: October 2, 2006