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BIOCONTROL OF ROOT ROT OF AVOCADO SEEDLINGS

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ABSTRACT

Microbial isolates previously reported to suppress root rot of blue lupin seedlings (after inoculation with Phytophthora cinnamomi [Pc]) were evaluated for suppression of Pc root rot of avocado seedlings.

Three fungal isolates (Aspergilluscandidus, Paecilomyces lilacinus and Trichoderma hamatum) *and two bacterial isolates* (Bacillus azotoformans *and B.* megatenum) *reduced Pc root rot development and root colonisation of avocado seedlings (Cv. Edranol) significantly.*

UITTREKSEL

Isolate van mikro-organismes voorheen gerapporteer om wortelvrot van blou Iupiensaailinge (na inokulasie met Phytophthora cinnamomi [Pc]) te onderdruk, is geévalueervironderdrukking van Pc-wortelvrot van avokadosaailinge.

Drie swamisolate Aspergillus candidus, Paecilomyces lilacinus en Trichoderma hamatumj en twee bakterie-isolate (Bacillus azotoformans en B. megaterium,) het Pcwortelvrot en -wortelkolonisasie van avokadosaailinge (Cv. Edranol) betekenisvol onderdruk.

INTRODUCTION

Soil suppressive to avocado root rot development (caused by *Phytophthora cinnamomi [Pc])* in Australia, were shown to be more fertile and to contain higher numbers of microorganisms, such as bacteria (eg. *Bacillus* spp.), actinomycetes (eg. *Streptomyces* spp.), and fungi (eg. *Aspergillus* spp., *Penicillium* spp., and *Trichoderma spp.)*, than soils conducive to root rot development (Broadbent & Baker, 1974; Malajczuk & Mc Comb, 1979; Weste & Vithanage, 1977). Microbial populations have been credited for the suppressive ability of these soils (Halsall, 1982; Malajczuk, 1979; 1983) and have been implicated for differences in disease severity in forest soils (Weste *et al.*, 1977) and Eucalyptus spp. (Malajczuk, McComb & Parker, 1977). However, little information is available concerning biological control of root rot by specific antagonists.

The purpose of this study was to evaluate micro-organisms reported previously to suppress *Pc* root rot of blue lupin seedlings (Duvenhage *et al.*, 1991), for suppression of

Pc root rot of avocado.

MATERIALS AND METHODS

Evaluations of antagonistic micro-organisms for the control of root rot of *Persea Americana* (Cv. Edranol) seedlings

Inoculum of antagonists and *Pc* was obtained and each antagonist used to inoculate nursery planting medium together with *Pc* as described by Duvenhage *et al.*, (1991).

The nursery planting medium consisted of 50% composted sugar cane residue and 50% sifted sand, which was sterilised by methyl bromide fumigation and adequately aerated before use.

Edranol seedlings were planted in the inoculated planting medium in 4ℓ plastic bags using 10 replicates per treatment. Bags were then placed in a nursery covered with 40% shadow netting and watered three times a week. Temperature fluctuated between 18 and 30°C.

Seedlings were removed from bags after three months and the roots were rinsed under running tap water. Root rot seventy was rated on a percentage scale as follows:

0= No visible sign of disease

- 1= Less than 20% root rot
- 2= 21-40% root rot
- 3= 41-60% root rot
- 4= 61 -80% root rot
- 5= More than 80% root rot

Colonisation of roots by *Pc* was determined by dipping feeder roots in 70% ethanol for 5s, blotting dry, cutting into small pieces (in the direction of the root tips), and plating 10 randomly selected pieces on each of 5 corn meal agar plates (Zentmyer, 1963). The number of root pieces yielding growth of *Pc* was recorded and expressed as the percentage root pieces colonised.

Cultures of fungi were deposited at the National Collection of Fungi (Plant Protection Research Institute, Pretoria) and identified. Cultures of bacteria and actinomycetes were identified by the Vegetable and Ornamental Plant Institute (Pretoria).

RESULTS

Evaluations of antagonistic micro-organisms for the control of root rot of *Persea Americana* (Cv. Edranol) seedlings

Of the 24 isolates of micro-organisms tested for the control of root rot of avocado seedlings, 2 isolates of bacteria (*Bacillus azotoformans* [B43] and *B. megaterium* [B42]) and 3 fungi (*Aspergillus candidus* [F44; PREM. 50935], *Paecilomyces lilacinus* [F40; PREM. 50933] and *Trichoderma hamatum* [F56; PREM. 50938]) reduced *Pc* root rot

and extent of root colonisation significantly when compared to the inoculated control treatments (Tables 1 and 2). No actinomycete reduced *Pc* root rot or extent of root colonisation significantly when compared to the inoculated control (Table 3).

 TABLE 1
 Effect of antagonistic bacteria on root rot of Edranol seedlings planted in Pc-inoculated nursery plant

 medium

Isolate code	* Root rot rating	# Percentage of root pieces colonised
B41	4,8 a	86 a
Inoculated control (without antagonist)	4,4 a	82 a
В3	4,2 ab	77 a
B5	4,1 abc	78 a
B42	3,4 bc	62 b
B43	3,3 c	62 b
Uninoculated control	0,0 d	0,0 c

Values not followed by the same letter are significantly different according to Duncan's multiple range test (P = 0,05).

- * Mean root rot rating of ten replicate avocado seedlings.
- # Mean percentage of five replicate plates with ten root pieces each, yielding growth of Pc.
- **TABLE 2** Effect of antagonistic fungi on root rot of Edranol seedlings planted in *Pc*-inoculated nursery plant medium

Isolate code	* Root rot rating	# Percentage of root pieces colonised
Inoculated control (without antagonist)	4,4 a	84 a
F42	3,9 ab	71 ab
F46	3,6 b	70 ab
F56	3,4 b	65 b
F40	3,4 b	63 b
F44	3,3 b	58 b
Uninoculated control	0,0 c	0,0 c

Values not followed by the same letter are significantly different according to Duncan's multiple range test (P = 0.05).

* Mean root rot rating of ten replicate avocado seedlings.

Mean percentage of five replicate plates with ten root pieces each, yielding growth of Pc.

Isolate code	* Root rot rating	# Percentage of root pieces colonised
A5	4,9 a	88 ab
A11	4,9 a	89 ab
A17	4,9 a	92 a
A46	4,8 a	87 abc
Inoculated control (without antagonist)	4,1 b	82 bcd
A47	3,9 b	75 d
A1	3,8 b	78 cd
Uninoculated control	0,0 c	0,0 e

TABLE 3 Effect of antagonistic actinomycetes on root rot of Edranol seedlings planted in *Pc*- inoculated nursery plant medium

Values not followed by the same letter are significantly different according to Duncan's multiple range test (P = 0,05).

* Mean root rot rating of ten replicate avocado seedlings.

Mean percentage of five replicate plates with ten root pieces each, yielding growth of Pc.

DISCUSSION

Antagonist species found to reduce root rot of avocado in this study (*Bacillus* spp. and *Trichoderma* spp.) are known antagoniststo *Phytophthora* spp. (Malajczuk, 1983), and high numbers of *Aspergillus* spp. have previously been associated with suppressive soils (Malacjzuk *et al.*, 1979). However, no previous reports of effective root rot control of avocado by the microbial species reported here as effective biocontrol agents, could be found. Results obtained in this study show that control of *Pc* root rot of avocado can be obtained by application of single antagonist species as biocontrol agents in a commercially used nursery medium.

REFERENCES

- BROADBENT, P. & BAKER, K.F. 1974. Behaviour of *Phytophthora cinnamomi* in soils suppressive and conducive to root rot. *Australian Journal of Agricultural Research* 25: 121 137.
- BROADBENT, P., BAKER, K.F. & WATERWORTH, Y. 1971. Bacteria and actinomycetes antagonistic to fungal root pathogens in Australian soils. *Australian Journal of Biological Science* 24: 925 944.
- DUVENHAGE, J.A., KOTZÉ, J.M. & MAAS, E.M.C. 1991. Suppressive soils and biological control of *Phytophthora* root rot. *South African Avocado Growers' Association Yearbook* 14: 6 11.
- HALSALL, D.M. 1982. A forest soil suppressive to *Phytophthora cinnamomi* and conducive to *Phytophthora cryptogea*. II Suppression of sporulation. *Australian Journal of Botany* 30: 27 37.
- MALAJCZUK, N. 1979. Biological suppression of *Phytophthora cinnamomi* in Eucalypts and avocados in Australia. Pages 635 - 652 in: *Soil-borne Plant Pathogens.* Eds. Schippers, B. & Gams, W.

MALAJCZUK, N., 1983. Microbial antagonism to Phytophthora. Pages 197 - 218 in:

Phytophthora: It's biology, taxonomy, ecology and pathology. Eds., Erwin, D.C., Bartnicki-Garcia, S. & Tsao, P.H., 392 pages.

- MALAJCZUK, N. & McCOMB, A.J. 1979. The microflora of unsuberized roots of Eucalyptus calophylla R. Br. and Eucalyptus marginata Donn ex Sm. seedlings grown in soil suppressive and conducive to Phytophthora cinnamomi Bands. I Rhizosphere Bacteria, Actinomycetes and Fungi. Australian Journal of Botany 27: 235 - 254.
- MALAJCZUK, N., McCOMB, A.J. & PARKER, C.A. 1977. Infection by *Phytophthora cinnamomi* Rands of roots of *Eucalyptus calophylla* R. Br. and *Eucalyptus marginata* Donn. ex Sm. *Australian Journal of Botany* 25: 483 500.
- WESTE, G. & VITHANAGE.K. 1977. Microbial populations of three forest soils: seasonal variations and changes associated with *Phytophthora cinnamomi*. *Australian Journal of Botany* 25: 377 383.
- ZENTMYER, G.A., 1963. Biological control of *Phytophthora* root rot of avocado with alfalfa meal. *Phytopathology* 53, 12: 1383 1387.