

SENSITIVITY OF FUNGAL PATHOGENS TO CHEMICAL SUBSTANCES IN AVOCADO TREES

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OPSOMMING

Metanol ekstrakte van die blare, mesokarp, sade, skille en wortels afkomstig van sewe verskillende avokadobome is getoets vir antifungiese aktiwiteit teenoor ses wortelpatogene en ses na-oespatogene, asook teenoor twee swamme wat nie patogenias vir avokado's is nie. Geen konsekwente verskille in antifungiese werking het voorgekom tussen die verskillende bome nie. Op 'n gemiddelde basis was die aktiwiteit van die blaarekstrakte die hoogste, gevolg deur die mesokarp-, wortel-, saaden skillekstrakte respektiewelik.

SUMMARY

Methanol extracts of the leaves, mesocarp, seeds, skins and roots from seven different avocado trees were tested for antifungal activity towards six root pathogens and six post-harvest pathogens, as well as two fungi non-pathogenic to avocado. Differences in activity between trees were inconsistent. On average, the extracts of leaf tissue exhibited the greatest fungitoxicity, followed by the mesocarp-, root-, seed- and skin extracts respectively.

INTRODUCTION

There is increasing evidence that fungitoxic chemical substances play a role in the resistance of some avocado trees to infection by *Phytophthora cinnamomi*. Zaki *et al.*, (1973) reported the presence of a potent preformed antifungal agent in tissues of *Persea* spp resistant to *P. cinnamomi*, but not in susceptible species. This chemical compound was later given the name borbonol (Zaki *et al.*, 1980). Dixon and van Lelyveld (1981) found five fungitoxic compounds to occur at similar levels in the seedling roots of avocado cultivars Duke (moderately resistant to *P. cinnamomi*) and Fuerte (moderately susceptible). The fungitoxic chemical substances in avocado tissue do not seem to be specific for *P. cinnamomi*, since the presence of the substances is determined by bio-assaying with *Cladosporium cucumerinum* (Zaki *et al.*, 1980) and a *Penicillium* sp (Dixon and van Lelyveld, 1981).

Wehner and Apostolides (1981) have previously reported differences in fungitoxicity towards avocado pathogens of chemical substances extracted from avocado cultivars Edranol, Duke 6 and Duke 7. In this paper evidence is presented of differences in fungitoxicity of extracts from different parts of various avocado trees. Some of these trees varied in susceptibility to *P. cinnamomi*.

MATERIALS AND METHODS

Leaf, fruit and root samples of seven avocado trees were collected at Westfalia Estate, Duiwelskloof (Table 1). The fruits were peeled and the seeds were removed. Leaf, seed, skin, mesocarp and root samples were extracted with methanol (500 ml methanol/100 g plant material) by homogenization. The extracts were filtered, dried with anhydrous CaSO₄ and concentrated in vacuo at 70°C to a volume of ca 50 ml.

Whatman antibiotic assay discs, 13 mm in diameter, were impregnated with the extracts (300 µl extract/disc) and the solvent evaporated at 70°C. Petridishes containing malt extract agar were inoculated centrally with the following fungi. *Phytophthora cinnamomi*, *P. cinnamomi* R4C (isolated from a tree treated with Ridomil for four years), *Rhizoctonia solani*, *Cylindrocarpon destructans*, *Fusarium oxysporum* (two isolates), *Thyronectria pseudotrichia*, *Colletotrichum gloeosporioides*, *Dothiorella aromática*, *Fusarium solani*, *Lasiodiplodia theobromae* (previously known as *Botryodiplodia theobromae*), *Phomopsis perseae*, *Drechslera sorokiniana* and *Phytophthora citrophthora*. The extract impregnated discs were placed in duplicate on the agar in the Petridishes at equal distances from the inocula (5 discs/petridish). After incubation for up to 10 d at 25°C, the effect on fungal growth was determined according to the following scale.

0. stimulation
1. no effect
- 2: inhibition after fungus growth has reached disc
- 3: slight inhibition (reduction in mycelium density)
- 4: inhibition
- 5: pronounced inhibition

TABLE 2: Effect of methanol extracts of avocado tissue on the growth of fungi

Plant part	Tree No.	Rate of inhibition													
		<u>P. cinnamomi</u>	<u>P. cinnamomi R4C</u>	<u>R. solani</u>	<u>C. destructans</u>	<u>F. oxysporum 2</u>	<u>F. oxysporum 3</u>	<u>T. pseudotrithia</u>	<u>C. gloeosporioides</u>	<u>D. aromatica</u>	<u>F. solani</u>	<u>L. theobromae</u>	<u>P. perseae</u>	<u>D. sorokiniana</u>	<u>P. citrophthora</u>
Leaf	1	3	3	2	1	0	0	0	0	5	3	3	4	3	3
	2	3	3	2	1	0	0	0	3	4	3	4	4	3	1
	3	3	3	0	1	1	0	0	4	4	3	5	4	3	3
	4	3	3	0	1	0	0	0	1	4	3	5	4	3	3
	5	1	3	0	1	1	0	0	5	4	1	5	4	3	3
	6	3	3	0	1	0	0	0	5	4	3	5	5	3	3
	7	1	3	0	3	4	0	0	5	3	4	5	5	3	3
	Mean	2,4	3,0	0,6	1,3	0,9	0	0	3,3	4,0	2,9	4,6	4,3	3,0	2,7
Seed	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	2	4	3	2	1	0	0	0	0	4	3	1	1	4	1
	3	1	3	2	1	1	1	0	1	3	3	1	1	3	4
	4	3	3	0	1	0	0	0	1	4	3	1	0	3	3
	5	1	3	1	1	1	0	0	1	4	3	1	3	3	3
	6	1	1	1	1	1	1	1	1	1	1	1	1	1	1
	7	4	4	0	1	1	0	0:3	1	4	3	1	3	3	3
	Mean	2,3	2,8	1,0	1,0	0,7	0,3	0,4	0,8	3,3	2,7	1,0	1,5	2,8	2,5
Skin	1	1	1	2	3	1	1	1	1	4	3	1	1	1	1
	2	1	3	2	1	1	1	0	1	3	3	1	1	3	4
	3	1	1	0	3	1	1	3	5	3	1	0	1	1	1
	4	1	1	0	1	1	1	0	1	4	1	1	0	3	3
	5	0	1	3	1	1	1	0	3	3	3	1	1	3	3
	6	1	1	0	3	1	1	1	3	1	1	1	0	1	1
	7	0	1	0	1	1	1	0	1	3	1	1	1	3	1
	Mean	0,7	1,3	1,0	1,9	1,0	1,0	0,7	2,1	3,0	1,9	0,9	0,7	2,1	2,0
Mesocarp	1	3	1	3	1	1	1	1	3	1	1	1	1	1	0
	2	3	3	3	3	3	1	4	5	1	1	1	3	1	1
	3	3	3	3	1	1	1	5	4	3	1	1	0	1	1
	4	3	3	3	1	1	1	5	4	3	1	1	3	1	1
	5	3	3	0	1	1	1	5	4	1	1	1	3	1	1
	6	1	3	3	3	3	1	5	4	1	3	1	3	1	1
	7	3	3	3	1	1	1	3	4	1	1	1	3	1	1
	Mean	2,7	2,7	2,6	1,6	1,6	1,0	4,0	4,0	1,6	1,3	1,0	2,3	1,0	0,9
Root	1	0	3	2	1	0	1	0:4	0	4	3	0	2	3	0:3
	2	4	4	2	1	1	1	0	1	4	1	1	1	3	1
	3	4	3	0	1	1	1	0:3	1	1	1	1	1	3	3
	4	4	3	0	1	0	0	0:3	1	5	1	1	0	3	4
	5	4	4	0	1	1	0	0:3	1	4	1	1	3	3	3
	6	4	4	0	1	0	0	0:3	1	0	3	1	1	3	3
	7	4	4	0	1	0	0	0	1	5	3	1	1	4	3
	Mean	3,4	3,6	0,6	1,0	0,4	0,4	1,2	0,9	3,3	1,9	0,9	1,3	3,1	2,6

RESULTS

In 40% of the cases, the extracts had no effect on fungus growth, while stimulation occurred in 17%. Pronounced inhibition (rating 5) was observed in 4% of the cases, inhibition (rating 4) in 10% and slight inhibition (rating 3) in 27% (Table 2). In some instances, inhibition as well as stimulation was observed. A considerable degree of variation was evident in the fungitoxicity of the various extracts towards the different fungi. However, there were no consistent tendencies observed in the fungitoxicity of the extracts from the various trees. Neither did the different scions or the trees with different disease ratings differ from each other in fungitoxicity. The most pronounced differences were observed between extracts from different plant parts. Extracts of leaf tissue exhibited the greatest fungitoxicity, followed by mesocarp, root, seed and skin extracts. The growth of fungi non-pathogenic to avocado viz. *D. sorokiniana* and *P. citrophthora* was also inhibited by many of the extracts.

Of special interest is the fact that, on average, post-harvest pathogens were inhibited to a considerably higher degree by extracts from leaves than were root pathogens. This difference was not evident in the case of seed, skin, mesocarp and root extracts. Root extracts had the greatest inhibitory effect on both *P. cinnamomi* isolates used in this study. The rate of inhibition of *P. cinnamomi* R4C was generally slightly greater than in the case of the "wild type". *T. pseudotrichia* was inhibited to a considerable degree by mesocarp extracts but not to such an extent by the other extracts, while *L. theobromae* was inhibited only by leaf extracts. *F. solani* was consistently inhibited more than its sister species *F. oxysporum*.

DISCUSSION

It is clear that fungitoxic chemical substances were present in all the trees examined. The results obtained further indicate that the fungi-toxicity observed could be ascribed to more than one compound and that these compounds occurred in different parts of the plant, rather than in different trees. However, Zaki et al., (1980) readily extracted and detected the antifungal compound borbonol in leaves, stems and roots of *Persea* spp. Similarly, Bergman and Beijersbergen (1968) isolated the fungitoxic lactone, α -methylene butyrolactone in considerable quantities from all parts of the tulip plants examined by them. The stimulatory effect observed in some instances could probably be ascribed to the presence of nutrients in the extracts. Where fungitoxic substances were also present, stimulation as well as inhibition occurred.

The results of this study forms a basis for further investigations. It is envisaged to determine the oligodynamic effect of the fungitoxic substances and to establish the nature of the compounds. Since the trees from which the samples were taken are available for further studies, the in vivo resistance to the various pathogens could be determined and the results compared with those of this in vitro survey.

TABLE 1: Description of avocado trees investigated

No.		Description
1	Fuerte/Duke	Escapee
2	Ryan/Mexican	Escapee
3	Fuerte/Guatemala	Escapee
4	Fuerte/Guatemala	Disease rating 3
5	Fuerte/Guatemala	Disease rating 6
6	Fuerte/Duke	Disease rating 3
7	Fuerte/Duke	Disease rating 6

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