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Susceptibility of *Pinus oocarpa* to *Armillaria* root disease in Zimbabwe

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The results of susceptibilities to *Armillaria heimii* Pegler of 30 eight to ten year old *Pinus oocarpa* clones, with up to 56 ramets each, in five seed orchards are presented. Deaths among clones ranged from nil to 9.36 percent during the four-year study period. Average annual mortalities in seed orchards were 1.20 percent to 5.23 percent. There were statistically significant differences among the clones within and across seed orchards. Duncan's multiple range comparisons of proportions of dead trees in each clone indicated that clones of South African origin were generally least susceptible. Clones from Nicaragua were most susceptible. There was a significant negative linear progression of symptoms and positive progression of deaths in clones during the study period.

Keywords: *Armillaria heimii*, *P. oocarpa*, clones, Zimbabwe.

Introduction

In Zimbabwe over 70 percent of the industrial plantation area estimated at 100 000 hectares is planted to *Pinus* species. *P. elliottii*, *P. patula* and *P. taeda* account for over 90 percent of the area planted to pines (Forestry Commission, 1989). The area planted to *Pinus oocarpa* is about 2 000 hectares. The species is one of the most drought tolerant pines at low altitudes (<1 000 m.a.s.l.), but is sensitive to frost (Barnes, 1993) and is preferentially uprooted by the warthog (*Phacochoerus aethiopicus*). *P. oocarpa* is susceptible to *Armillaria* root disease (Masuka, 1989). The current and postulated buoyant demand for local timber and timber products has led to the afforestation of climatically and physiographically marginal sites. The area planted to *P. oocarpa* and other lesser known pine species is, therefore, increasing.

Armillaria species are soil-borne pathogens which, under favourable conditions, invade the cambium and sapwood of the roots and stem, forming diagnostic mycelial fans under the bark (Gibson, 1975). They are endemic in Zimbabwe in indigenous forests dominated by *Brachystegia spiciformis*, *Bridelia micrantha*, *Harungana madagascariensis*, *Parinari*

curatelifolia and *Vernonia* species, causing non-lethal infections and deaths (Masuka, 1993). *Armillaria* has caused serious losses in susceptible pine species in Zimbabwe since systematic afforestation was initiated in the early 1960s. This prompted studies on the occurrence, distribution and ecology of the fungus, predisposition to attack and differential susceptibility of pines (Swift, 1968; 1972). Localised deaths can be as high as 25 percent but *Armillaria*-induced mortalities in plantations are generally regarded as being insignificant (Masuka, 1989; 1993). Recent studies on the taxonomy of *Armillaria* species in Zimbabwe have shown the existence of three morphological and molecular groups, including *A. heimii* (Pegler) and *A. mellea* (Vahl) Fr. (Mwenje and Ride, 1993).

Worldwide, *Armillaria* species have caused losses in forest trees and the genus has been a subject of recent intensive investigations (Shaw and Kile, 1991).

Intraspecies differences in susceptibility to the fungus have not been studied in pine species grown in Zimbabwe. The present study sought to investigate clonal variation in *P. oocarpa* in susceptibility to *Armillaria* in several seed orchards at one site.

Materials and Methods

Trial site and history

The five seed orchards are all at Mukandi, John Meikle Forest Research Station (1 760 m.a.s.l., 18°44'S and 32°41'). The orchards were planted between 1983 and 1985. The major indigenous tree species before the area was planted to *P. oocarpa* were *Parinari curatelifolia*, *Harungana madagascariensis*, *Cussonia natalensis*, *Bridelia micrantha*, *Albizia gumifera*, *Catha edulis* and *Trema orientalis*. This vegetation was cut, stumped, stacked and burnt before planting positions were made out at an espacement of 7 x 7 m. Clones (cleft-grafted onto a single local rootstock) were randomly allocated to rows in a seed orchard. Thirty clones (see Table 2) with up to 50 ramets each were planted in a seed orchard. The species collected from infected indigenous and pine trees at the locality is *Armillaria heimii* Pegler (*Armillaria* Project Report, 1992).

Assessment for Armillaria

The procedure for the detection of *Armillaria*-induced deaths followed that outlined in Masuka (1989) thus: inspecting for mycelial fans under the bark by exposing the root collar and major roots; examining for mycelial fans between the bark and wood of the stem; and inspecting for resin exudation on the stem and root collar region of some major lateral roots. Yellowing and resinosis

in trees are generally indicative of *Armillaria* infection at the site (Masuka, 1989). The analyses of variance (ANOVA) and regressions for the proportions of infected and dead ramets in clones were based on arcsine-transformed data. Duncan's multiple comparison tests were based on transformed and un-transformed proportions.

Results

Average annual mortalities in seed orchards were 1.20 percent to 5.23 percent during the four year period (Figure 1). Deaths among clones ranged from nil to 9.36 percent (see Table 2). Initially, seed orchards were analysed separately at each assessment interval. There were significant differences ($p < 0.05$) in the proportions of deaths due to *A. heimii* among the 30 clones during the four year period in all seed orchards except SO81 (Table 1). The differences in the proportions of symptomatic trees among clones were also significant ($p < 0.05$) across all seed orchards. Seasonal differences (= years) significantly ($p < 0.01$) influenced the infection pattern in seed orchards SO79 and SO82. Variation in the proportions of deaths among clones was only insignificant ($p > 0.05$) in seed orchard SO69. The results, generally, indicate a strong influence of the performance of ramets by scions.

Table 1: Analysis of variance for deaths due to *Armillaria* among seed orchards.

Seed orchard	No. of ramets (1988)	Mean Square			
		Clone		Year	
		infected	dead	infected	dead
69	321	0.0170**	0.0121**	0.0024ns	0.0002ns
79	1300	0.0480**	0.0276**	0.1012**	0.0470**
80	1013	0.0140**	0.0104**	0.0046ns	0.0285**
81	412	0.0199**	0.0084ns	0.0124ns	0.0225*
82	367	0.0337**	0.209**	0.0780**	0.0305*

Sns not significant, * significant at $p < 0.05$, and ** significant at $p < 0.01$.

Duncan's multiple comparison test, based on transformed and un-transformed data, revealed that all clones, except 94 and 944 for deaths, and 943 and 944 for symptoms, were significantly different ($p < 0.05$) during the study period (Table 2). The number of dead and symptomatic trees strongly depended ($p < 0.001$) on geographical origin of the clones. Clones of Nicaraguan and Honduran origin were most susceptible, while those of South African were least susceptible to *A. heimi* (Table 2). The results presented are based on

un-transformed data. There was considerable variation in the performance of clones with a similar geographic origin (country). Honduran clones 946 and 947 had the highest and 944 least proportions of dead ramets. Most clones from South Africa exhibited moderate to high tolerance to *A. heimi*. Clones such as 622 (from South Africa) and 1 122 (from Guatemala) had a high proportion of infected ramets and a low proportion of dead ramets. There was, however, a general consistency among clones and geographical origin in the rankings for both proportions of dead and symptomatic trees (Table 2).

Table 2: Origin of clones and Duncan's ranking of untransformed means.

Clone	Origin	Proportion Rank	Dead	Rank	Symptomatic
946	Honduras	1	0.0936	7	0.0470
947	Honduras	2	0.0887	4	0.0924
1123	Nicaragua	3	0.0656	3	0.1373
942	Guatemala	4	0.0655	6	0.0619
1127	Nicaragua	5	0.0633	5	0.0777
90	South Africa	6	0.0392	8	0.0420
1125	Guatemala	7	0.0392	10	0.0393
93	South Africa	8	0.0332	16	0.0199
623	South Africa	9	0.0295	11	0.0319
945	Honduras	10	0.0241	17	0.0167
1129	Guatemala	11	0.0210	23	0.0066
624	South Africa	12	0.0209	26	0.0031
1126	Honduras	13	0.0161	24	0.0051
622	South Africa	14	0.0159	2	0.1373
620	South Africa	15	0.0153	9	0.0410
1130	Honduras	16	0.0142	15	0.0213
940	Guatemala	17	0.0139	30	0.0005
621	South Africa	18	0.0126	27	0.0029
943	Guatemala	19	0.1150	29	0.0015b
1124	Honduras	20	0.1150	13	0.0235
91	South Africa	21	0.0108	25	0.0042
941	Guatemala	22	0.0106	12	0.0264
1122	Guatemala	23	0.0096	1	0.1403
1131	unknown	24	0.0083	21	0.0113
625	South Africa	25	0.0064	19	0.0131
92	South Africa	26	0.0054	20	0.0115
619	South Africa	27	0.0028	14	0.0224
1128	Guatemala	28	0.0016	22	0.0067
94	South Africa	29	0.0000a	18	0.0135
944	Honduras	30	0.0000a	28	0.0015b
	Nicaragua	1	0.0661	1	0.1086
	Honduras	2	0.0369	3	0.0312
	Guatemala	3	0.0223	2	0.0327
	South Africa	4	0.0161	4	0.0278
	Unknown	5	0.0092	5	0.0120

Clones 90 and 1125 and 943 and 1124 have the same proportions of dead ramets due to rounding-off to four decimal figures. Proportions followed by the same letter are not significantly different at $p < 0.05$.

The general response pattern of clones during the study period was investigated by pooling proportions of dead or symptomatic ramets in each clone across seed orchards. There was a linear decrease in the proportions of symptomatic trees (-0.02 ; $p < 0.01$) and linear increase in proportions of dead trees (0.01 ; $p < 0.01$) (Table 3). The response of orchards \times year: linear was non-significant ($p > 0.05$) for the proportions of dead ramets (Table 4).

P. chiapensis and *P. patula* seed orchards, despite having the same history, and similar planting and management practices. A previous report (Masuka, 1989) had indicated that there were some clones with consistently higher mortalities, irrespective of the seed orchard and position within an orchard.

There have been very few reports on rootstock influence on disease resistance (Jayawickrama *et al.*, 1991), but Hollis *et al.*, (1979) found that the rootstock had an effect on the susceptibility of *P. elliottii* ramets to rust. In the present study, all the rootstock material was from a single seed-lot (half-sib material). The results may therefore, be

Table 3: Pooled regression coefficients for linear and quadratic contrasts.

Variable	Linear contrast	Regression	
		Linear	Quadratic
Dead	0.10**	0.01**	0.0002 ^{ns}
Symptomatic	-0.17**	-0.02**	-0.004**

ns not significant; ** significant at $p = 0.01$

Table 4: Pooled single class regression analyses for the proportions of dead and symptomatic ramets.

Source	DF	MS	
		Dead	Symptomatic
Year:Linear	1	0.1410***	0.1974**
Clones	29	0.0436***	0.0825***
Clones \times Year:linear	29	0.0139***	0.0282***
Orchards	4	0.0681***	0.1562***
Orchards \times Year:Linear	4	0.0059 ^{ns}	0.0327***
Residual	437	0.0056	0.0071

ns not significant; ** significant at $p = 0.01$; *** significant at $p = 0.001$

Discussion

Ramet influence on rootstock performance

The *P. taeda* rootstock was used because it is known, from past experience, to be less susceptible than *P. oocarpa* rootstock to warthog damage. Assessment for *Armillaria* root disease in the seed orchards was prompted by the high mortality observed in some clones in the orchards, compared to adjacent *P. taeda* (even of the same rootstock),

regarded as reflecting true variation among clones as ramets from a single source material have the same set of disease resistance genes (Zobel and Talbert, 1984).

Disease progression

Repeated measurements were made on the same trees during the four year period. The observed differences among clones are those due to treatment effects at each assessment

(Rowell and Walters, 1976). There is a general indication that susceptibility to attack decreases with age, though already infected trees may succumb to the fungus, resulting in the continued upward linear trend in the proportions of dead trees among clones (Table 3). Under local conditions the resistance of pines to attack by *Armillaria* increases after eight years (Swift, 1972) and decreases after about 18 years depending on the species (Masuka, 1993). The non-significant linear regression between orchards for the proportion of dead ramets in each clone might have been confounded with the significant orchard x year interaction (Table 4). The non-significant single class regression among orchards indicates that some seed orchards were more susceptible than others (Table 4). Seasonal variation, especially in the amount and distribution of rainfall might predispose trees to attack or accelerate the deaths of those already infected. Precipitation and temperature are the major determinants of both latitudinal and altitudinal distributions of *Armillaria* (Hood *et al.*, 1991). In Zimbabwe, as elsewhere in tropical Africa, most deaths due to *Armillaria* are confined to cool, moist and high elevations formerly occupied by indigenous tree species.

Clonal forestry can be preferred in tree improvement because it provides an opportunity to utilise genotype variation directly (Leakey, 1987). The use of clonal material enables a differentiation and evaluation of the importance of stress and vigour in infection by a fungus. A distinction of the effects of host susceptibility and *Armillaria* ecology has been reported to be difficult (Shaw and Kile, 1991), especially if the experimental material is limited, as both may limit the opportunities for contact between the fungus and potential hosts. In the present study over 3 000 trees in five seed orchards were assessed. The material can, therefore, be regarded as sufficient to indicate real trends in susceptibility/tolerance among clones. The effect of environment on the observed responses among clones could not be evaluated as the trial was at one site only. Clones showing promise, such as the majority

of those of South African origin, are recommended for use in advanced generation tree improvement studies on the species.

The importance of *A. heimii*, and other *Armillaria* species, in pine plantations might be under-estimated. An assessment of the loss due to *Armillaria* should include aspects of mortality, loss of growth increment in infected trees and the value of land out of production due to high mortalities.

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