Effects of Mycosphaerella Leaf Spot on Growth of *Eucalyptus nitens*

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ABSTRACT

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Standard disease assessment diagrams for estimating the severity of Mycosphaerella leaf spot, caused by *Mycosphaerella nubilosa*, were developed based on percentage of spotted leaf area and degree of defoliation. The diagrams were used to evaluate disease severity in 11 provenances of *Eucalyptus nitens* growing in Eastern Transvaal, South Africa. Variation in disease severity was found among and within provenances. Provenances from New South Wales, Australia, were more resistant than those from Victoria. Growth rate was negatively correlated with disease severity but was not affected until 25% of the crown was defoliated.

Nearly 50% of the commercial forestry area in South Africa is planted to *Eucalyptus* spp. (3). Of this area, 78% (287,649 ha) is planted to *Eucalyptus grandis* Hill ex Maiden. Because land suitable to grow *E. grandis* is becoming scarce and expensive, forest growers are investigating the possibility of using more readily available and less expensive high-altitude sites on the Transvaal Highveld. However, *E. grandis* cannot grow on the highveld because of frequent frosts. *E. nitens* (Deane et Maiden) Maiden is a desirable alternative species because it is frost-tolerant, grows rapidly, and produces wood on short rotation suitable for mining timbers and pulpwood (16). Because *E. nitens* produces seed sparingly and seed years are irregular, seed is expensive and often difficult to obtain. Most seed, in fact, must be imported to South Africa from Victoria (VIC) and New South Wales (NSW), Australia, because few local seed stands exist. Provenance research of *E. nitens* in South Africa has been reviewed previously (2,15).

A potential obstacle to growing *E. nitens* is Mycosphaerella leaf spot (MLS), caused by *Mycosphaerella nubilosa* (Cke.) Hansf. This disease causes severe spotting and premature defoliation of juvenile leaves (13). Mature foliage is not infected. MLS was first reported in 1933 from an unspecified *Eucalyptus* sp. near Capetown (17), but records in the National Collection of Fungi, Pretoria, show that the disease had actually been discovered in 1925 at Hogsback State Forest in the eastern Cape Province on *E. globulus* Labill. (4). At that time, *E. globulus* was one of the most commonly planted commercial tree species in South Africa, but it was abandoned during the early 1930s. The abandonment has often been attributed to the eucalyptus snout beetle (*Gonipterus scutellatus* Gyllenhal), but in some areas, unpublished reports noted that MLS was so severe that the species was considered unsuitable for afforestation (9). No records of the disease appeared again until 1980 (10), when it was reported in a *E. nitens* provenance trial (11), where NSW provenances were noted to be more resistant than VIC provenances. Since then, MLS has been found throughout the highveld areas of Southeastern Transvaal and Northern Natal and has been one of the most frequently reported diseases of *Eucalyptus* spp. in South Africa (8).

The purpose of this study was to design a useful method for measuring MLS in the field and to use this method to assess the effects of disease on growth of NSW and VIC provenances of *E. nitens* growing at a single location in South Africa.

MATERIALS AND METHODS

Study site. Eleven provenances of *E. nitens* from VIC and NSW were planted in a trial during March 1980 at the Jessievale State Forest (26° 14′ S latitude, 30° 31′ E longitude, 1,733 m altitude) on the Eastern Transvaal Highveld (Table 1). The trial was planted in a randomized complete block design with between two and five replicates of 16 trees planted in square plots at a spacing of 2.7 × 2.7 m. Uneven replicates were necessary because of inadequate quantities of seed from certain provenances.

Disease assessment. To determine the pattern of lesions occurring on individual leaves, infected juvenile leaves were analyzed visually for lesion pattern, size, and shape. Generally, lesions had a random pattern over the leaf surface, and were mostly 1–1.5 mm in diameter and angular in shape. Based on these observations, a series of disease assessment diagrams (DAD) were drawn to represent leaves with 3, 6, 12, 25, 50, and 75% symptomatic surface area (Fig. 1). A logarithmic scale was used because it allows accurate visual estimates of disease intensity (1,6). Disease severity was

Table 1. Estimates of disease severity of 11 *Eucalyptus nitens* provenances using defoliation score and percent leaf spot

CSIRO[a] stock number	Provenance[b]	Defoliation score (rank) 1983	Defoliation score (rank) 1984	Percent leaf spot
9471	Ebor (NSW)	1.8 (1)	5.6 (1)	23.6
12121	Tallaganda (NSW)	1.8 (1)	5.7 (3)	25.8
11162	Nimmitabel (NSW)	3.0 (3)	5.6 (1)	32.4
12120	S. Anembo Trig. (NSW)	3.3 (4)	5.7 (3)	26.6
8414	Barrington Tops (NSW)	4.0 (5)	6.1 (7)	26.0
12114	Braidwood (NSW)	4.3 (6)	5.9 (6)	28.5
9514	Parker's Gap (NSW)	4.3 (6)	5.8 (5)	32.9
12211	Alexandra (VIC)	6.0 (8)	6.6 (8)	35.3
12175	Toorongo Plateau (VIC)	6.2 (9)	7.3 (11)	42.6
12102	Noojee (VIC)	6.3 (10)	6.9 (9)	39.1
12107	NW Traralgon (VIC)	6.4 (11)	6.9 (9)	38.1
Mean		4.30	6.20	31.90
F (provenances)		19.64**[c]	12.02**	10.91**
NSW mean		3.20	5.80	27.90
VIC mean		6.20	6.90	38.80
t (NSW, VIC mean)		7.24**	6.99**	6.89**

[a] Commonwealth Scientific and Industrial Research Organization, Division of Forest Research, Canberra, ACT, Australia.
[b] NSW = New South Wales and VIC = Victoria, Australia.
[c] ** = Significant at $P < 0.01$.

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subsequently determined by standing within 2 m of the crown of each tree, observing leaves in the crown at or near breast height to develop an impression of their average disease condition, and estimating the average percentage of leaf surface with lesions with the aid of the DAD.

To determine the pattern of symptoms that occurred in the crowns of individual trees, 20 trees showing a wide range of disease severity were selected, and each was measured at height intervals of 0.5 m for width of defoliated region, width of region with spotted foliage, and total crown width. Generally, defoliation occurred at the lower and inner part of the crown and extended progressively upward in a cone in more heavily infected trees. Surrounding the defoliation column was a zone of diseased foliage surrounded by asymptomatic foliage.

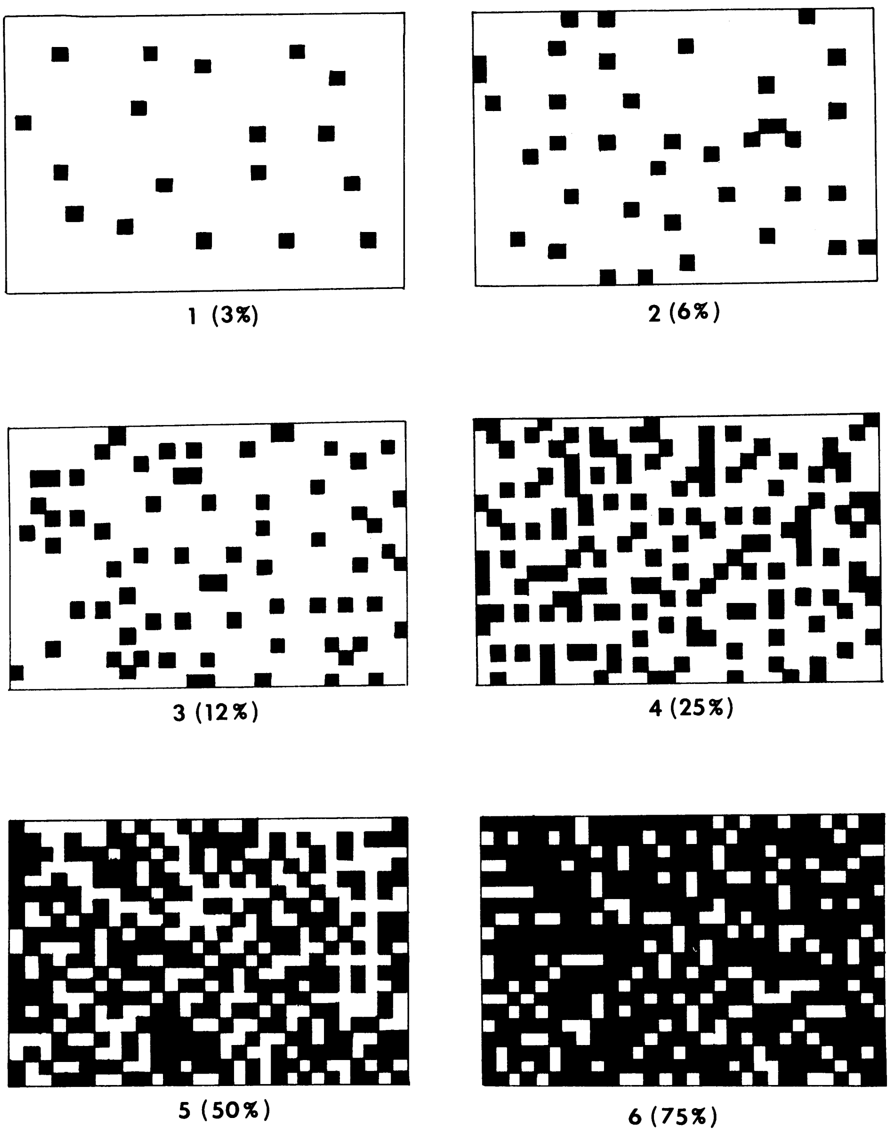

Fig. 1. Disease assessment diagrams based on amount of spotted leaf area used to rate disease severity of Mycosphaerella leaf spot on 3- and 4-yr-old *Eucalyptus nitens.* Solid black regions represent disease lesions. Actual leaf size varies according to provenance. Leaves sampled averaged 5–10 cm wide and 20–25 cm long.

Table 2. Analysis of variance of defoliation scores in 1983 and 1984

Source	df	1983 SS[a]	1983 Percent of total SS	1984 SS	1984 Percent of total SS
Replicate (R)	3	34.12	1.3	25.44	2.8
Provenance (P)	10	1,549.31	57.7	193.05	20.8
R × P	25	197.17	7.4	40.16	4.3
Within P	538	902.90	33.6	668.13	72.1

[a] Sums of squares.

Based on these observations, a second series of DAD was drawn to represent trees showing 0, 3, 6, 12, 38, 50, 63, and 75% defoliated crown area (Fig. 2). Defoliation severity was subsequently determined by standing 2–5 m from each tree and matching crown condition to the standard diagrams and assigning it a disease score: 1 = 0–3%, 2 = >3–6%, 3 = >6–12%, 4 = >12–25%, 5 = >25–38%, 6 = >38–50%, 7 = >50–63%, 8 = >63–75%, and 9 = >75%.

Measurements based on leaf spot percentage and defoliation score were made in February 1983. These measurements showed that leaf spot percentage and defoliation score were highly correlated (Table 1, $r = 0.99$, $P < 0.01$). Because defoliation was assessed more easily and was highly correlated with leaf spot percentage, only defoliation was assessed in February 1984. On both dates of disease measurement, juvenile foliage was present on all trees of all provenances and no crown closure had occurred. By 1985, most trees had primarily mature foliage and, consequently, too little disease for further assessment.

Growth assessment. Diameter at breast height and total height of each tree were measured in February 1983, February 1984, and March 1986. Individual tree volumes were calculated by the following formula developed by Opie (12):

$$VOL = (DBH^2\,HT)/10^{(4.762-(5613/(DBH+127)^2))},$$

where VOL = underbark volume in cubic meters, DBH = overbark diameter at breast height in centimeters, and HT = height in meters. To calculate growth increments for the 1983/1984 and 1984/1986 seasons, total volume, diameter, and height for 1983 and 1984 were subtracted from total volume, diameter, and height for 1984 and 1986, respectively.

RESULTS

Disease assessment. Differences among provenances for defoliation scores in 1983 and 1984 were significant ($P < 0.01$), and the NSW provenances consistently had lower defoliation scores than the VIC provenances (Table 1). The respective average defoliation scores for VIC and NSW provenances were 6.2 and 3.2 in 1983 and 6.9 and 5.8 in 1984 and were significantly different ($P < 0.01$) in both years. There was also considerable variation among individuals within most provenances for defoliation score for both years (Table 2). Within-provenance variation accounted for 33.6 and 72.1% of the total variation in 1983 and 1984, respectively. Individual trees in some VIC provenances had low defoliation scores, which were comparable to trees in the NSW provenances.

Growth assessment. Defoliation scores were used to examine the relationship between growth and disease severity. Analyses were based on average growth of all trees within each defoliation score. Based on both 1983 and 1984 scores, volume, height, and diameter increments during the 1983/1984 season all showed highly significant curvilinear relationships with disease severity for that year (Fig. 3). With the 1983 scores, growth was not negatively affected until reaching a score of 5 (>25–38%). With the 1984 scores, no trees showed a defoliation score below 4 (>12–25%), above which growth in diameter and volume were progressively less with progressively higher scores. Growth in height was not affected until a 1984 score of 6 (>38–50%), above which it showed the same response trend as diameter and volume.

Using the regression models developed from the 1984 scores, percentage losses in volume, diameter, and height were calculated for increasing levels of defoliation (Table 3). MLS had more of an impact on diameter than height until reaching a defoliation score of 8, after which height was affected more. At all scores above 4, volume growth was affected more than diameter or height growth.

Individual trees varied both in diameter at breast height and height at the beginning of the observation period. To determine how disease affected growth of trees in different initial size classes, trees were grouped into five classes according to initial (1983) diameters (class 1 = <3.5 cm, 2 = 3.6–5.5 cm, 3 = 5.6–7.5 cm, 4 = 7.6–9.5 cm, and 5 = >9.5 cm) and initial heights (class 1 = <3.5 m, 2 = 3.6–5.5 m, 3 = 5.6–7.5 m, 4 = 7.6–9.5 m, and 5 = >9.5 m). Because so few trees fell within height classes 1 and 2, only classes 3, 4, and 5 were used for analyses. Generally, trees in the higher initial size classes grew more rapidly than those in the smaller size classes for each defoliation score. Within each size class, there was a range of defoliation scores and a general downward

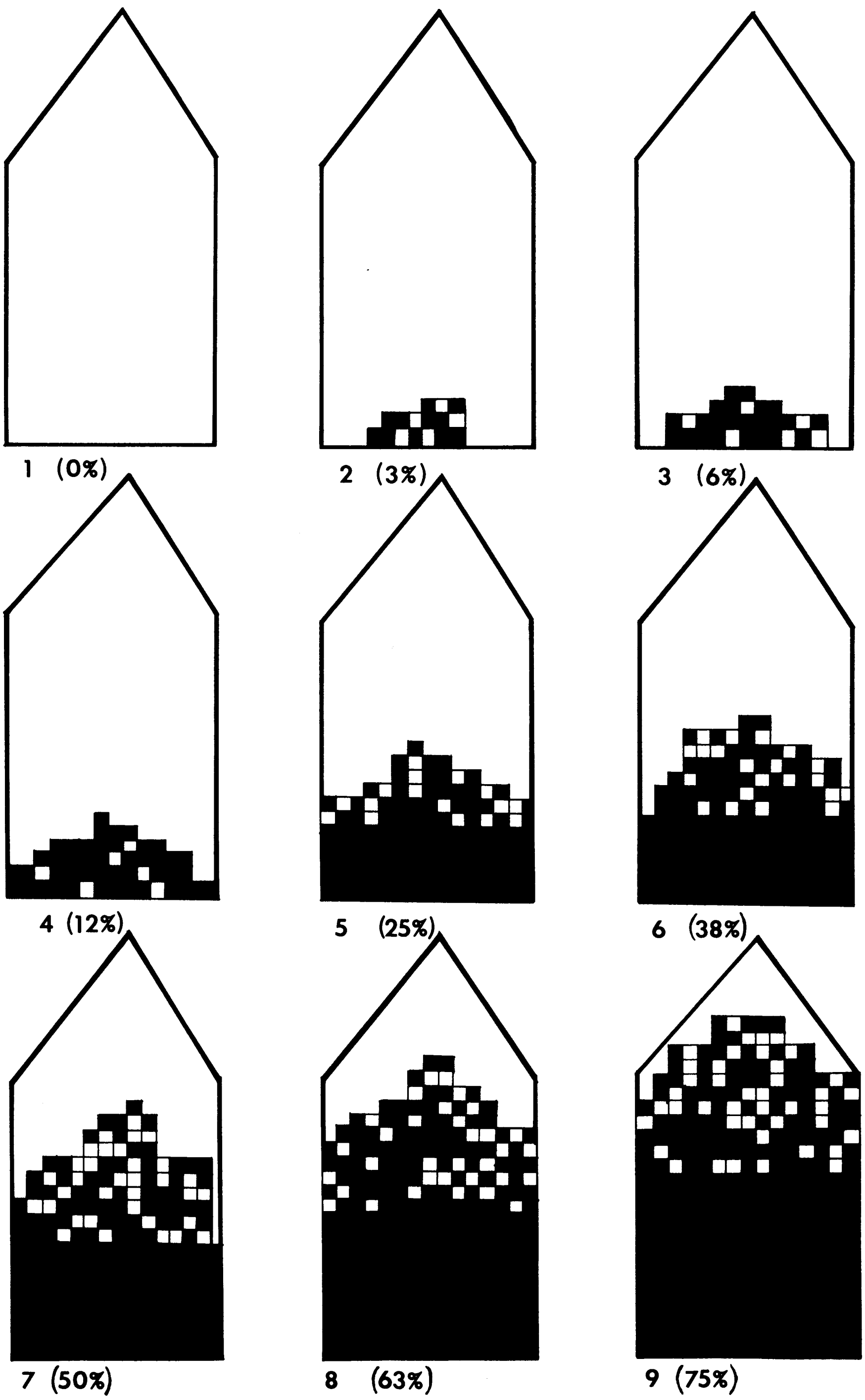

Fig. 2. Disease assessment diagrams based on amount of defoliation in crown used to rate disease severity of Mycosphaerella leaf spot of 3- and 4-yr-old *Eucalyptus nitens*. Solid black regions represent zones of complete defoliation, and partially black regions represent zones of partial defoliation. Lower edges of diagrams represent bases of the trees.

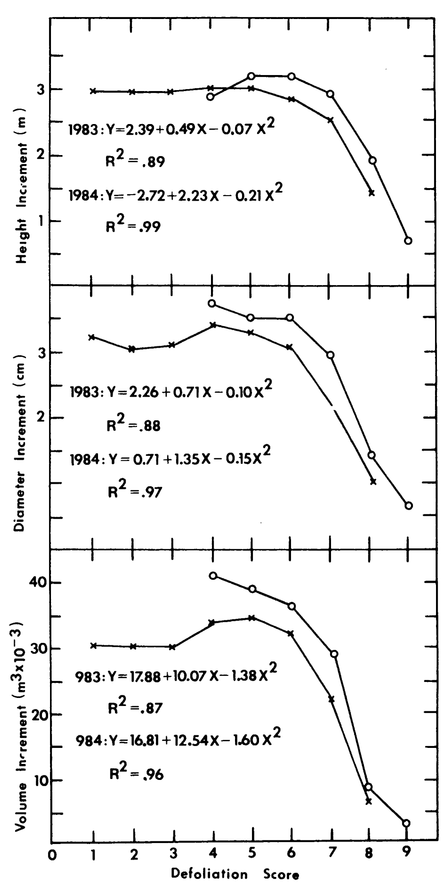

Fig. 3. Effects of severity of Mycosphaerella leaf spot on height, diameter, and volume increments on infected 3-yr-old (X———X) and 4-yr-old (o———o) *Eucalyptus nitens*. Defoliation scores: 1 = 0–3%, 2 = >3–6%, 3 = >6–12%, 4 = >12–25%, 5 = >25–38%, 6 = >38–50%, 7 = >50–63%, 8 = >63–75%, and 9 = >75%.

trend in growth as defoliation score increased (Fig. 4).

A comparison of growth responses between NSW combined provenances and VIC combined provenances indicated both sources responded similarly to increasing defoliation in terms of decreasing volume growth (Fig. 5). A similar relationship was found with height and diameter growth. In a related study (15), the averages for NSW combined provenances for total volume, height, and diameter were 40, 14, and 20% greater than those for VIC combined provenances.

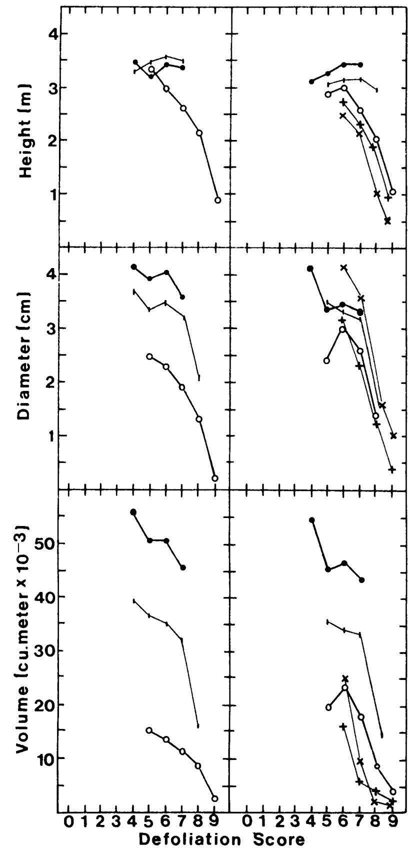

Fig. 4. Effects of severity of Mycosphaerella leaf spot on height, diameter, and volume increments on infected 4-yr-old *Eucalyptus nitens* according to diameter and height classes. For diameter classes (left column), (×——×) = <3.5 cm, +——+ = 3.6–5.5 cm, o——o = 5.6–7.5 cm, /——/ = 7.6–9.5 cm, and •——• = >9.5 cm. For height classes (right column), o——o = 5.6–7.5 m, /——/ = 7.6–9.5 m, and •——• = >9.5 m.

During the postinfection period (1984–1986), height, diameter, and volume increments for both VIC and NSW all showed downward trends with increasing 1984 defoliation score. When trees were grouped according to initial size classes, no consistent relationship with defoliation score was noted, although some classes did show downward trends with increasing defoliation score (Fig. 6). Trees with larger initial sizes grew more rapidly than smaller trees.

DISCUSSION

The series of disease assessment diagrams representing severity of leaf spotting and defoliation were found useful for field rating of Mycosphaerella leaf spot in *Eucalyptus* trees. Measurements based on both these parameters were significantly correlated with each other and could quantitatively define differences among provenances and individual trees. Measurements based on defoliation were quicker and easier to make; however, trees in this study were measured in late summer, when symptoms were well advanced. If measurements were taken earlier in the summer, the leaf spotting symptom would probably be the more useful measure. Furthermore, disease severity was measured only once during each season. Because disease severity varies throughout the year and the rate at which it changes can differ between years, scores cannot be directly compared between years. In this study, for instance, trees assessed in 1984 showed generally higher average scores and greater within-provenance variation than in 1983 even though measurements were made at precisely the same time of the year. This indicates that conditions for disease development were probably more favorable in 1984 than 1983. Despite the variation between years, the relative rankings of provenances remained nearly the same.

Pederick (14) showed that *E. nitens* exhibits large variation among prove-

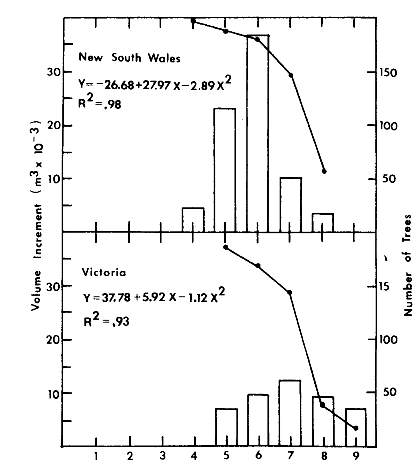

Fig. 5. Effects of severity of Mycosphaerella leaf spot on volume increments (1983–1984) on 4-yr-old *Eucalyptus nitens* trees originating from New South Wales and Victoria, Australia. Bar chart refers to number of infected trees within each defoliation zone.

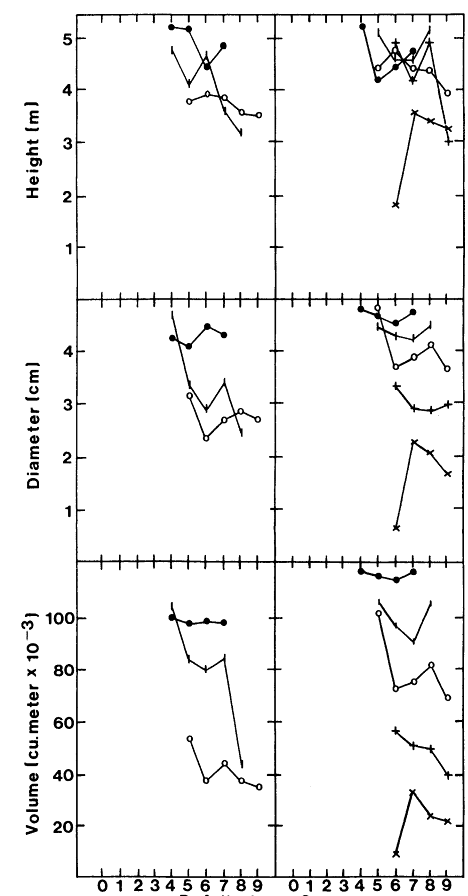

Fig. 6. Height, diameter, and volume increments summed for fifth and sixth years of growth of *Eucalyptus nitens* according to 1983 diameter and height classes and 1984 defoliation scores. For diameter classes (left column), ×——× = <3.5 cm, +——+ = 3.6–5.5 cm, o——o = 5.6–7.5 cm, /——/ = 7.6–9.5 cm, and •——• = >9.5 cm. For height classes (right column), o——o = 5.6–7.5 m, /——/ = 7.6–9.5 m, and •——• = >9.5m.

Table 3. Growth loss associated with different levels of defoliation of 4-yr-old *Eucalyptus nitens* caused by Mycosphaerella leaf spot calculated by the regression equations based on 1984 disease severity vs. growth assessments

	Percent loss of potential growth increment		
Defoliation score	**Height**	**Diameter**	**Volume**
1 (0–3%)	0	0	0
2 (>3–6%)	0	0	0
3 (>6–12%)	0	0	0
4 (>12–25%)	0	0	0
5 (>25–38%)	0	0	4
6 (>38–50%)	2	8	17
7 (>50–63%)	18	24	37
8 (>63–75%)	47	48	64
9 (>75%)	91	81	99

nances in ability to retain juvenile foliage, and those provenances that retain their juvenile foliage longer have greater growth rates. By causing a premature loss of juvenile leaves, Mycosphaerella leaf spot could be restricting growth. In this study, whether the differences in size of trees at 3 yr were due to a history of disease is impossible to determine, because measurements were not taken during the first 3 yr of growth. Nevertheless, trees within each size class showed an inverse relationship with defoliation score during the fourth year. During the postinfection period between 4 and 6 yr, no such relationship was evident, even though smaller trees continued to grow less rapidly. The lasting effect of disease was to cause a reduction in growth, and trees that became suppressed did not subsequently grow as rapidly as larger trees. Furthermore, the evidence presented here indicates that Mycosphaerella leaf spot causes a reduction in growth rate in *E. nitens* when more than 25% of the juvenile crown is defoliated. Similar trends have associated with Dothistroma blight on *Pinus radiata* D. Don in Kenya, where the critical point was 20% defoliation (5), and are well-known from artificial and insect defoliation studies (7).

On the basis of the 1984 scores, both NSW and VIC provenances showed similar responses to disease. NSW provenances, however, had an overall average volume of 40% more than VIC. The data presented here suggest that, at the same severity of infection, there is no difference between the growth responses of the NSW and VIC provenances. There is evidence, however, that most of the trees from the NSW provenances had infection severities below the point at which Mycosphaerella leaf spot severely affected growth, indicating greater resistance to the disease.

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