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Armillaria root disease of *Pinus radiata* in New Zealand (4)

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ARMILLARIA ROOT DISEASE OF *PINUS RADIATA* IN NEW ZEALAND: 4. ASSESSMENT OF STAND INFECTION

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ABSTRACT

Levels of infestation by *Armillaria* species are less readily ascertained in many newer plantations of *Pinus radiata* D. Don in New Zealand, because early mortality is generally lower than when stands were planted directly on ex-indigenous forest sites. Two methods of assessing infection were therefore explored using data from a second-rotation trial for which infection was known for every tree. Theoretical transects of different sizes placed randomly in this stand determined that single-tree transects were the most efficient, requiring examination of the least number of root collars for the same level of accuracy. The numbers of trees were determined that should be sampled to achieve required levels of precision for different intensities of infection. Incidence and severity of infection at age 6 years were found to be greater nearer to trees killed early in the rotation period, suggesting an alternative assessment approach. Computer-generated contour maps of the incidence of killed and living infected trees were used to demonstrate that field maps of the distribution of visible, low-intensity, pre-thinning mortality may have potential for identifying sites with greater overall stand infection. This method could simplify the operational evaluation of stand infection, but more field work is required to establish its feasibility.

Keywords: root disease; assessment; incidence of infection; *Armillaria novae-zelandiae*; *Armillaria limonea*; *Pinus radiata*.

INTRODUCTION

The root disease fungi *Armillaria novae-zelandiae* (Stevenson) Herink and *A. limonea* (Stevenson) Boesewinkel are widespread in New Zealand exotic forests, particularly in plantations of *Pinus radiata* (Hood 1989; Self *et al*. 1998). Due to the altered, less conspicuous nature of the disease, fresh approaches are needed for distinguishing stands with higher levels of infection. Previously, the wide distribution of obvious early mortality clearly identified the worst areas. Data from a second-rotation trial, in which root collar infection had been determined on every tree, were used to explore alternative options for estimating the distribution and incidence of infection. Two approaches were considered: direct assessment of stand infection from root collar inspections of a sample of standing trees, and indirect estimation derived from a record of the distribution and incidence of early mortality. The first procedure had already been used on a number of occasions, but the comprehensive

data now available made possible an examination of the method and precision of sampling. The second approach was likely to be more cost-efficient, if its estimate of stand infection proved reliable, since even low-incidence early-mortality is clearly visible and more readily assessed. The potential value of this option was explored by investigating a possible relationship between the distributions of *Armillaria* species in living and dead trees.

MATERIAL AND METHODS

Field Data

Data were gathered from a 3.1-ha trial in Kaingaroa Forest, established to determine the effect of thinning and pruning on the incidence of infection by *Armillaria* species, mainly *A. novae-zelandiae*, in second-rotation *P. radiata* on a non-indigenous forest site. The stand has been described fully by Hood & Sandberg (1993) and Hood *et al*. (2002). Trees were planted at an average density of 810 stems/ha in 1985 after harvesting of a first-rotation crop of 54-year-old *Pinus nigra* ssp. *laricio* (Poir.) Maire. The positions of all trees in the Kaingaroa trial were mapped prior to crown closure, and data were collected at age 6 years, before the first thinning and pruning treatments had been carried out. Severity of infection was evaluated on all standing trees by using a light, short-handled grubber to expose the root collar in order to estimate the extent of girdling by a zone of resinosis, accompanied by the presence of rhizomorphs. Trees were assigned to one of five girdling classes (0 = 0%; 1 = 1–25%; 2 = 26–50%; 3 = 51–100%; 4 = dead, with *Armillaria* species). This technique is believed to give a reliable estimate of true infection of *P. radiata* by species of *Armillaria* in New Zealand (Hood *et al*. 2002).

Analyses

Two methods of estimating the incidence of infection in a stand were investigated. The first requires the direct inspection of the root collars of a sample of trees within the stand. The second relies on an assessment of early mortality to give an indirect appraisal of the distribution and incidence of infected living trees.

The sampling design for the direct approach was examined using a SAS procedure (SAS Institute Inc. 1990) to place linear transects containing various numbers of trees randomly along tree lines in the Kaingaroa trial at age 6 years. The minimum numbers of transects of various lengths required to estimate the incidence of infected trees within different tolerance levels were determined.

The indirect technique was investigated by first examining the data to see if there was a spatial association between the distributions of dead trees and living infected trees. This was done by assigning grid co-ordinates to all trees in order to test the relationship between the local incidence of early mortality and the incidence and severity of infection in adjacent living trees. Mean percentage incidence of infection and mean girdling score were calculated for all living trees within a given radius of each dead tree, for a range of radii between 5 m and 70 m, and compared to the overall incidence and severity. Contour maps were then plotted of the distributions of proportions of trees killed or chronically infected. These maps were produced using the interpolation function in S-PLUS (SPLUS 1997). Comparisons were made between the distributions of killed and infected trees.

RESULTS

Stand infection levels assessed by placing random transects are shown in Fig. 1 and 2. Use of a transect only 10 trees long gave a result nearly as precise as one 40 trees long, for the inspection of fewer trees (Fig. 1). For instance, to estimate the level of infection within 30%

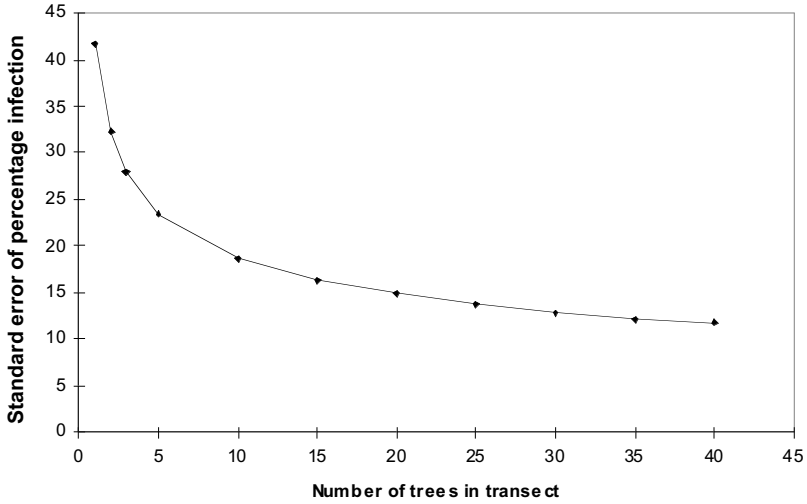

FIG. 1–Transect sampling simulation using unthinned Kaingaroa trial data: standard error for transects of different sizes

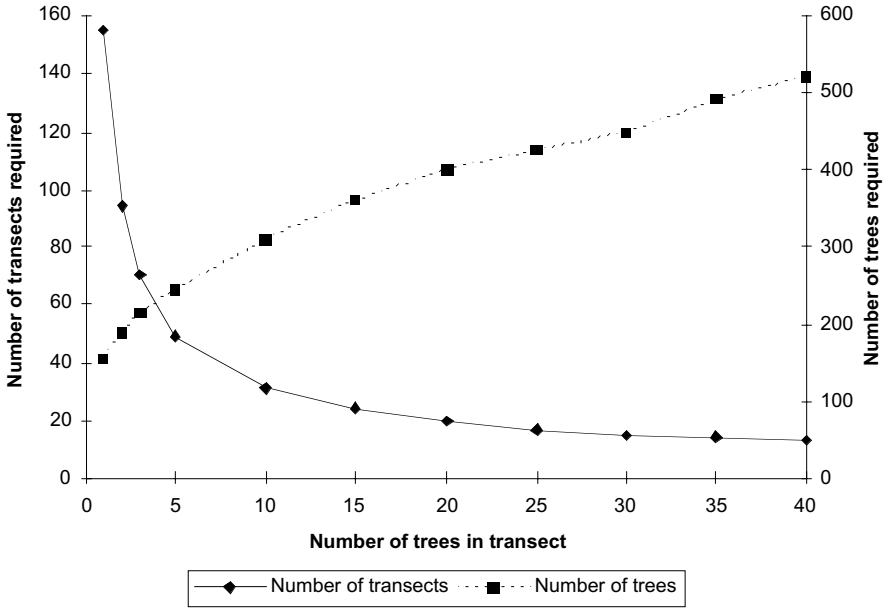

FIG. 2–Transect sampling simulation using unthinned Kaingaroa trial data: numbers of transects and numbers of trees required to determine infection to within 30% of the mean for transects of different sizes.

tolerance with 95% confidence (22±7% in the Kaingaroa trial at age 5 years) required 31 transects (310 trees) for a 10-tree transect, and ca. 13 transects (520 trees) for a 40-tree transect (Fig. 2). However, the most efficient of all was a one-tree transect, which required the inspection of only 150 trees to achieve a precision within 30% of the mean (Fig. 2). This is equivalent to 48 trees/ha, in the Kaingaroa trial, or a 6% sample in a stand stocked at 850 stems/ha. A more general result derived from binomial tables for single-tree sampling is shown in Table 1, which gives the precision of the mean percentage infection in a stand according to the number of trees sampled (sample size) and the underlying infection rate of the stand.

TABLE 1–The precision of mean percentage of trees infected by *Armillaria* species, obtained using individual tree sampling

Infection level	Sample mean percentage infection	Sample size				
		50	100	150	200	300
Low	4	5.5	4.4	3.5	3.0	2.4
Medium	13	10.7	7.0	5.7	4.9	4.0
High	30	13.3	9.4	7.6	6.6	5.3
Very high	70	13.3	9.4	7.6	6.6	5.3

The precision is expressed as a confidence interval. The population mean will be within plus or minus the tabulated value with 95% confidence. For example, if 4% of a 50-tree sample are infected, the underlying population infection rate will be approximately within 4±5.5% or 0%–10%.

Trees killed by *Armillaria* species early in the rotation were generally surrounded by a greater incidence of more severely infected living trees than occurred elsewhere in the stand, during the first evaluation (Fig. 3 and 4). Computer-derived contour maps of the local incidence of trees killed and infected by *Armillaria* species at age 6 years in the Kaingaroa

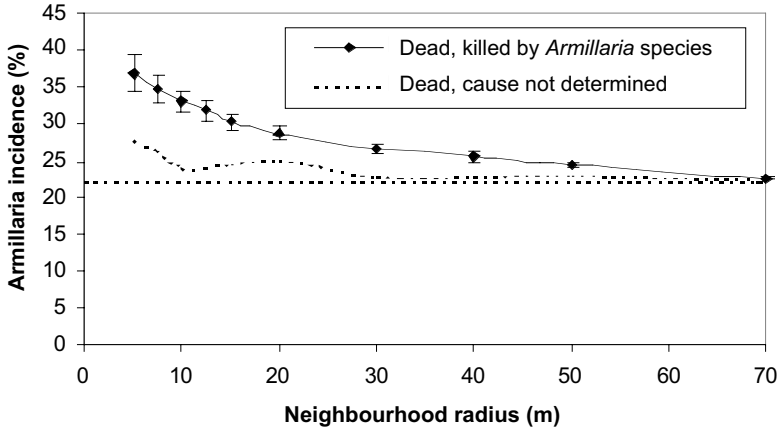

FIG. 3–Mean percentage incidence of living trees infected by *Armillaria* species at age 6 years, prior to thinning, within a given radius of trees that had died (error bars indicate standard errors; the horizontal broken line indicates the average stand incidence, independent of distance from dead trees).

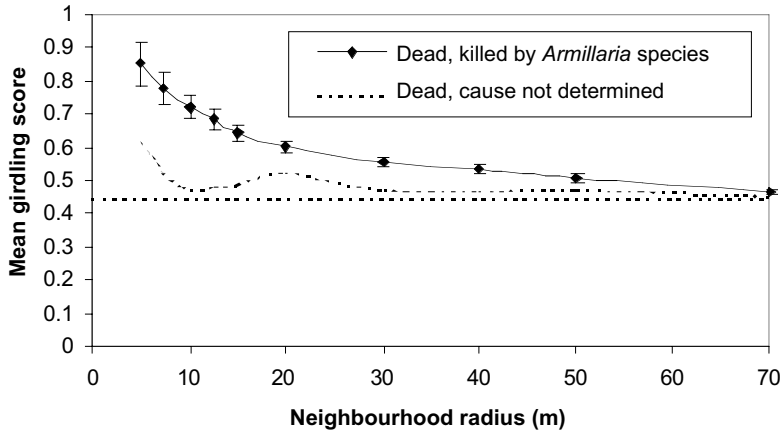

FIG. 4–Mean percentage severity of living trees infected by *Armillaria* species at age 6 years, prior to thinning, within a given radius of trees that had died (error bars indicate standard errors; the horizontal broken line indicates the average stand score, independent of distance from dead trees; maximum possible score, i.e., every tree completely girdled, 3.0).

trial are shown, respectively, in Fig. 5 and 6. There is a loose correlation between the distributions of dead and living infected trees (a correlation coefficient of 0.68 was obtained using a 4 × 4 grid of 16 plot means; $p<0.01$). In the Kaingaroa trial, areas of the stand where incidence of dead trees exceeded 3% approximated to areas where infection incidence in living trees exceeded 25%.

DISCUSSION

Recent estimates of growth loss due to Armillaria root disease indicate that it is valuable to be able to identify stands in which more than ca. 20% of apparently healthy trees are actually infected by species of *Armillaria* (Kimberley *et al*. 2002). The occurrence of such infection is variable, but indicated in broad terms by region and previous vegetation (Hood 1989; Self *et al*. 1998). In a recent countrywide survey, Self *et al*. (1998) found *Armillaria* species in 43 of 74 apparently healthy stands, at levels between 1% and 85% of trees infected. In the worst regions (Coromandel, the central North Island, Nelson, the South Island West Coast, and Southland), the mean incidence varied between 14% and 22%, and on ex-indigenous forest sites across the country, averaged 38%. Among second-rotation plantations, means ranged 10–20% on sites originally covered in indigenous forest or scrubland, but were lower on ex-herbaceous locations such as farmland. These means do not portray the extremes of variation between particular sites. In addition, incidence may be enhanced by host stress induced by management aspects (e.g., pruning, thinning, use of stock with imperfect root form, application of effluent waste, certain types of ground preparation, previous crop tree species) or influence of other disease organisms (e.g., *Dothistroma pini* Hulbary (Shaw & Toes 1977), and possibly *Cyclaneusma minus* (Butin) DiCosmo *et al*. and *Sphaeropsis sapinea* (Fr: Fr) Dyko & Sutton). The variation in incidence in regions and forests where *Armillaria* species are more likely to occur indicates a need for an ability to assess infection in specific stands and stand types.

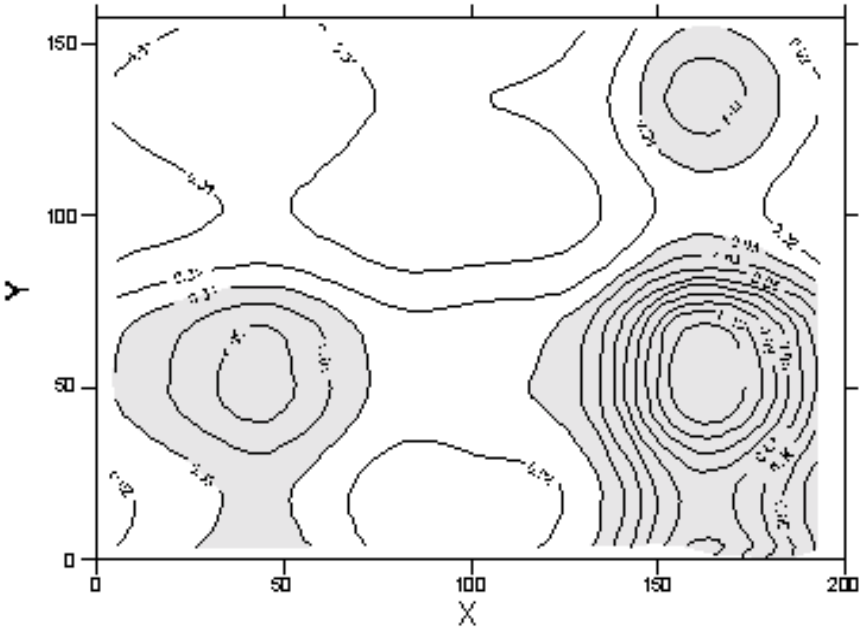

FIG. 5–Contour plot showing proportions of trees killed at age 6 years in the Kaingaroa trial (shaded area with > 3% of trees killed). Scale: co-ordinates of axes are in metres.

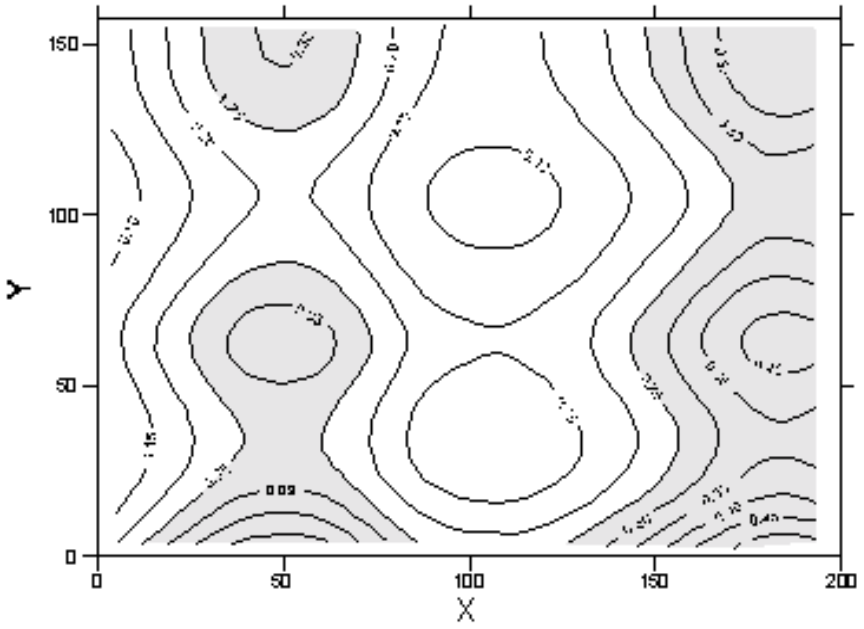

FIG. 6–Contour plot showing proportions of infected living trees at age 6 years in the Kaingaroa trial (shaded area with > 25% of trees infected). Scale: co-ordinates of axes are in metres.

Two methods of estimating stand infection were investigated. The technique of direct root collar examination has been refined for greater efficiency, and it is now possible to choose a sampling intensity that reflects the desired degree of precision according to the approximate level of infection anticipated. Precision depends on the number of trees sampled, not on the stand area. In practical terms it may be possible to stratify a forest according to zones of common vegetation history and past management, and assess a range of sample stands across each stratum. The unit area surveyed must be sampled representatively, and ideally at random (but probably in practice systematically). Costs for this method will be greater if topography is severe and access difficult, but efficiency may be improved by surveying prior to harvest, when slash is well decomposed, canopy closure has suppressed obstructing weed growth, and decisions regarding the next rotation can still be made. Alternately, sampling might be undertaken immediately prior to the first silvicultural operations, when management options are still open, the smaller tree size reduces root collar inspection time, and mortality may also be recorded, if comparison with the second method is desired.

The indirect approach, determining stand infection from visible mortality, was investigated by first testing the relationship between the distributions of infected trees and those killed by the pathogen, which was found to be significant. When mapped, the spatial occurrence of dead trees corresponded with the distribution of living infected trees (Fig. 5, 6). This distribution pattern remained stable at least through the first half of the rotation (Hood *et al*. 2002). The incidence of infection close to trees that were killed by *Armillaria* species was higher than the background incidence of 22% (Fig. 3). For example, trees within 5 m had an infection incidence of 37%. In Fig. 3 and 4, the curves for dead trees without symptoms of infection noticeably mirror those for trees where symptoms were present, for neighbourhood radii less than 10 m. Smaller trees killed by *Armillaria* species several years earlier were often dried and disintegrating, and such trees may not always retain clear evidence of infection (earliest mortality would date back to within 1 year of planting). The alternative explanation is much less likely in small trees, that possible colonisation by *Armillaria* species sometime after death could have generated sufficient inoculum potential to have influenced the incidence of infection in the neighbouring part of the stand (i.e., although linked, the association is probably non-causative).

The indirect approach therefore appears promising, but requires further investigation in different regions on a range of sites varying in incidence of infection. This method was investigated because, if effective, it appears potentially more cost-efficient than the direct method. Less time would be required to record the distribution of dead trees, whether undertaken aerially (Firth & Brownlie 2002) or from the ground, but either way a sample of trees would need inspection to verify cause of death. A mortality survey would also need to be timed within a very brief period of peak incidence, which may vary slightly in different stands (age ca. 4–6 years, before the initial thinning). Small plants that die earlier in the rotation would go undetected, even in a ground survey, and others would not have spread their roots sufficiently to encounter inoculum and become infected. On the other hand, once larger dead trees have been removed in the first thinning operation, there is usually little subsequent mortality. It may not be possible to use the survey information fully until after harvest, when delineated areas would be treated before the site is replanted with the next crop. However, the short rotation period of *P. radiata*, and the apparent constancy of spatial distribution of infection with time, make this a realistic procedure. Practically, a routine

survey would probably be conducted at a coarser scale of precision than that used in the Kaingaroa trial (Fig. 5, 6), since it seems unlikely that the extent of any remedial procedure would be less in area than a compartment or operational sub-compartment unit.

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