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What is the host range of *Phytophthora agathidicida* (kauri dieback disease) in New Zealand?

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Field Study: investigating the vegetation composition of kauri forest infected with *P. agathidicida*, versus kauri forest free from *P. agathidicida*



Figure.1 image showing crown dieback on kauri caused by *P. agathidicida*.

- Recce surveys were carried out within the Waitakere Ranges in West Auckland in infected and healthy kauri stands. Recording plant species abundances, along with many environmental characteristics.
- Ordination results showed a clear separation of infected versus healthy sites (Figure.2), shows infected sites are more similar in species composition to other infected sites and visa versa.
- Analysis of similarities shows that the difference in species composition between infected and healthy sites is significant (Figure.3)
- Whether this difference in species composition is due to direct impacts of the disease or indirect impacts due to the reduction of kauri needs to be investigated further.
- Environmental characteristics show only significant difference between healthy and infected sites pH and canopy density (Figure.4).

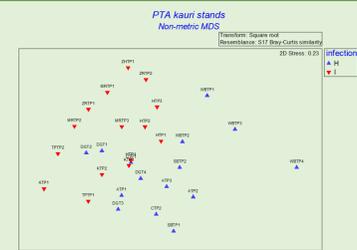


Figure.2 Ordination of infected (red) and healthy (blue) sites when similarity of species abundances at each site were compared.

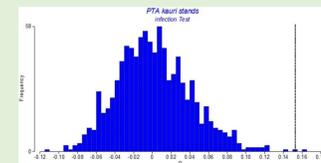


Figure.3 Analysis of similarities for the null hypothesis that there are no differences between healthy and infected sites. With a **R value equal to 0.153**, and a P value < 0.002.

| | mean (healthy sites) | mean (infected sites) | t-test significance value |
|---------------------------------|----------------------|-----------------------|---------------------------|
| mean canopy density (%) | 84.344 | 73.330 | 0.006685 |
| slope (°) | 28.333 | 28.733 | 0.937094 |
| altitude (m) | 173.467 | 146.733 | 0.147282 |
| East (aspect) | -0.01839233 | -0.342276625 | 0.221155 |
| North (aspect) | 0.184679415 | 0.043108262 | 0.582800 |
| soil pH | 4.8 | 5.3 | 0.003878 |
| soil total carbon (%) | 12.367 | 11.240 | 0.358177 |
| soil total nitrogen (%) | 0.497 | 0.515 | 0.628928 |
| soil total organic matter (%) | 21.29 | 19.40 | 0.369704 |
| soil Olsen phosphorus (mg/L) | 1.9 | 2.1 | 0.455498 |
| soil available nitrogen (kg/ha) | 189.3 | 198.7 | 0.573545 |
| kauri abundance (%) | 23.6 | 20.7 | 0.498416 |

Figure.4 Table of difference between environmental variables and the abundance of kauri in infected and healthy sites. Canopy density and soil pH are significantly different.

Detached-leaf assay: A quick method of studying the susceptibility of nine native plant species to *P. agathidicida*

- Leaves from nine native plant species including; *Knightia excelsa*, *Toronia toru*, *Leucopogon fasciculatus*, *Dracophyllum latifolium*, *Dracophyllum sinclairii*, *Astelia trinervia*, *Dianella nigra*, *Leionema nudum*, and *Phyllocladus trichomanoides* were collected from seedlings.
- An agar plug cultured with *P. agathidicida* was placed over small wound in the leaf surface and incubated in a tray, in the dark, at room temp for 13 days.
- Leaf tissue was then reisolated onto selective media to confirm Koch's postulates.

- Lesion growth occurred on *A. australis* (Figure.6), *D. latifolium*, *D. sinclairii*, *K. excelsa*, *L. nudum*, *L. fasciculatus*, and *T. toru* (Figure.5).
- Reisolation of *P. agathidicida* was seen from *A. australis* (Figure.7), *K. excelsa*, *L. fasciculatus* and *A. trinervia* infected leaves.

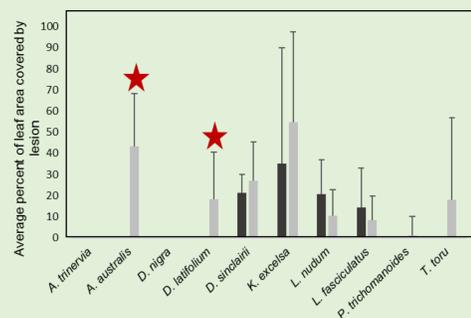


Figure.5 graph of average percentage of leaf surface covered by lesion. Red stars indicate a significant difference between control and inoculated tissue.



Figure.6 lesion growth on inoculated kauri leaves day 13

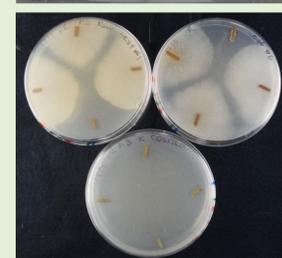


Figure.7 kauri re-isolation plates. Top two show inoculated leaf tissue growing *P. agathidicida*, bottom shows control leaf tissue with no pathogen growth.

Soil inoculation : Susceptibility testing for *P. agathidicida* on intact tanekaha and rewarewa seedlings

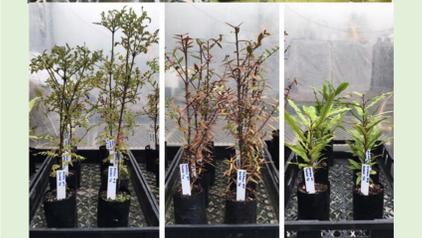


Figure. 6 shows inoculation of seedlings, (left to right) tanekaha, kauri, rewarewa



Figure. 7 shows seedling processing after 9 weeks, soil removal and scanning of roots.

- Soil inoculation of whole kauri, tanekaha, and rewarewa seedling was carried out (Figure.6).
- After 9 weeks growth seedlings were removed from the soil, and their roots scanned and analysed using WinRhizo software (Figure.7).
- Visual symptoms of infection were seen in both kauri and tanekaha, where the outside layer of the fine roots were nibbled away leaving only the inner vascular tissue (Figure.8).
- Neither tanekaha or rewarewa data show a significant impact by *P. agathidicida* according to WinRhizo output (Figure. 9)
- However, WinRhizo is known to struggle with picking up differences in very fine root tissues. Therefore with a longer trial period and younger seedlings it is possible that tanekaha or rewarewa would show a significant difference between inoculated and control roots.

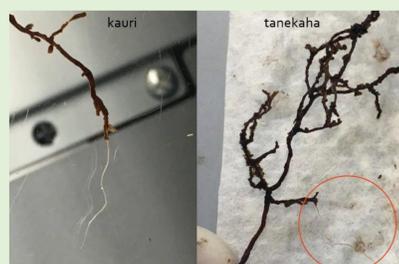


Figure. 8 shows seedling roots of kauri and tanekaha showing visual signs of *P. agathidicida* infection (nibbling of outer layer of root tissue)

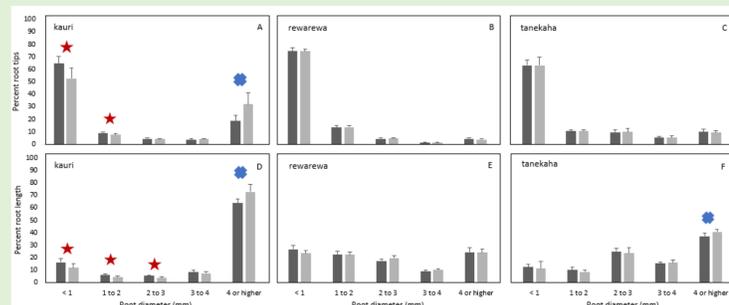


Figure. 9 shows graphs of number of root tips (A-C) and root length (D-F). Light grey bars show inoculated seedlings, Dark grey show control. Red star shows which diameter classes showed a significant difference where inoculated roots have less root tissue. Blue crosses show which diameter classes showed a significant difference where inoculated roots have more root tissue