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Phosphite Barriers for Kauri Dieback – Scoping Exercise

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EXECUTIVE SUMMARY

Phosphite Barriers for Kauri Dieback – Scoping Exercise

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July 2016

Kauri dieback, caused by *Phytophthora agathidicida*, is causing significant losses of kauri in New Zealand forests. Long-distance spread is mostly by human or large animal activity, but in infected stands there is a slow natural spread of the pathogen in the soil resulting from movement through the soil and via root-to-root contact. Over time, this gradual spread results in increasingly large diseased patches forming, potentially spreading over ridges and into neighbouring catchments.

This report outlines the potential for using phosphite barriers to halt or reduce the rate of spread of diseased patches of forest, and concludes that full-scale experimental investigation of phosphite barriers for kauri dieback control should be undertaken.

In theory, by treating a band of potential host species ahead of the disease front, those trees will be protected, thus forming a barrier preventing the proliferation of the pathogen and slowing its spread. In Australia, there is both experimental and anecdotal evidence that phosphite barriers slow the spread of dieback disease caused by *Phytophthora cinnamomi*. Phosphite barriers are being implemented and maintained on a large scale, particularly in Western Australia (WA). In New Zealand, phosphite has already been demonstrated to significantly reduce the growth of *P. agathidicida* in kauri trees. Although there is still some fine-tuning required regarding rates and application methods in kauri, it is now timely to extend research to investigate the effectiveness of phosphite deployment in barrier systems.

The barrier principle is accepted as sound by most researchers in the field, although most concede that gaining experimental proof-of-concept is challenging. This report reviews past and current work on phosphite barriers, identifies some of the factors that should be considered in research and proof of concept trials in kauri forest, recommends a logical progression of required work and assesses the feasibility of such work and barriers to success.

To justify any future deployment of phosphite barrier treatments in kauri forest, there needs to be proof-of-concept that the technique will be effective. This work must also determine whether there are any off-target effects of phosphite treatment, and to balance these against potential gains in kauri dieback control. Although the work will be long term, challenging and costly, such information gathering is essential to allow informed decision-making in the future.

The essence of any trials will be a comparison of the rate of pathogen spread in treated and untreated plots. Demonstration of a significant difference will be required for proof-of-concept. Demarcation of infection fronts before treatment application, and subsequent monitoring of infection front movement in treated and untreated plots in the years following treatment will be the primary measure for determining barrier effectiveness. To minimise the required number of samples and costs, the sampling will need to be done in a structured way over time to target the infection fronts. Soil testing using baiting techniques is currently the most reliable and accurate

way of determining *P. agathidicida* presence in the soil, although the project team should be prepared to adopt new and improved detection techniques if they are developed in the life of the trial.

Infection front demarcation and tracking will need to be backed up by kauri tree disease assessments, in particular canopy symptoms and basal trunk lesion development. Visual or higher tech options for such assessment could be used.

Ecological impacts of phosphite treatments, in particular immediate responses such as phytotoxicity, should be carefully monitored. This should include potential effects on mosses and lichens. In the longer term, potential effects such as differences in plant and microbial community composition in treated and non-treated, diseased and non-diseased areas should be assessed. There should be a focus on mycorrhizal associations to determine if phosphite has any effect on these. These ecological studies should be backed up by more intensive studies on other sites, to minimise disturbance and the potential for compromising the barrier trials.

Two different phosphite barrier treatment regimens will need to be tested alongside untreated control plots for comparison. The first is to treat only kauri, with trunk injection of trees and spray application of saplings or trees too small to inject. The second treatment regime is to treat all vegetation, including understorey and shrubs. Again, trunk injection should be used for trees, with spray application on plants too small to inject. The reason for this second treatment option is that the host range of *P. agathidicida* is not known. Alternative hosts could be harbouring or proliferating the pathogen without necessarily showing symptoms, potentially bridging the phosphite barrier if only kauri were treated. Having to include trees other than kauri considerably complicates the trials and increases the likelihood of off-target effects. But without this option, we could be in a situation in ten years' time where barriers might have failed and we don't know why. If there was good current knowledge of *P. agathidicida* hosts, this treatment could be modified or perhaps eliminated.

Current trials in kauri forests indicate that for trunk injection a phosphite concentration of 5% with 1 ml per cm trunk circumference is adequate and safe. By the time barrier treatment begins, more data should be available to confirm this. For spray application, the appropriate phosphite concentration is not known as work has not yet been carried out. A default concentration of 0.5% should be selected for spray application (this is the rate used in WA forest applications). It is recommended that spray application rates are investigated on common kauri-associated plants (including kauri) in advance of the treatment application in the barrier trials. This work should include investigation of appropriate surfactants required as spray additives.

The selection of suitable sites for the trial is a very important component of the programme. Some strict site criteria **must** be met, and other criteria will be desirable. The essential components of trial sites are: confirmed *P. agathidicida* presence, a contiguous 'clean' forest, sufficient area to allow at least one treated and one untreated control plot at each site, moderate to high stocking of kauri, and permission from forest owners/key stakeholders for long-term work on the site. These conditions cannot be compromised. Desirable but non-essential conditions include: flat to moderately sloped sites, good vector control, small (ricker) to moderate sized kauri trees, relatively easy site access (but not too public), and avoidance of other experiments or studies near the trial sites.

A number of different trial sites will be required. The variable nature of the bush and anticipated confounding factors expected in these trials means that at least six, and preferably 10 to 12 replicate plots of each treatment will be required. Plot size will need to be large, probably in the order of 20 x 30 m, possibly larger (depending on site factors such as topography and tree

size). Each site should have at least one treated and one untreated control plot. It is unlikely that there will be room for more than two or three replicates at any one site, with some sites only having one replicate. Although this adds to the complexity and logistical difficulties of the trial, it also strengthens the trial by testing in different soils, climates and vegetation types. In addition, it buffers against catastrophic events such as fire or flood, landowner revocation of permission, site interference and vandalism, and significant site disruption by vectors such as pigs.

There are a large number of potential trial sites, and as a starting point a majority of known kauri dieback sites could be considered to see if they meet the key criteria. But decisions on which sites will be most suitable cannot be made until sites are investigated in some detail, including soil sampling to determine if infection boundary demarcation is clear enough to be useful in the trial. Site selection, detailed mapping and demarcation of infections fronts will be a substantial body of work, probably taking many months. This work will be best done over the dry summer months, when site disturbance and spreading of disease will be minimised. There are likely to be sites where preliminary work is carried out, but sites don't end up in the barrier trial for various reasons. A likely situation is where infection boundaries cannot be found or delineated clearly enough for the trial requirements, but this will not be known until sampling and lab analyses have been done. Precise mapping will be required for the final selected sites, including all topographical, drainage and vegetation features, delineation of infection and disease boundaries, layout of plots and positions of barrier treatments.

Following the site sampling, final selection and infection front delineation stages, plots can be marked out and selected treatments applied. This will be followed by an intense period of monitoring for ecological impacts, focussing initially on determining if there are any phytotoxicity responses. Progression of disease symptoms in kauri should be assessed annually, and infection front progression should be re-mapped every 1–2 years, focussing on predetermined transects. Retreatment of plots will be required, probably after 3 to 5 years. More information on this will be available from other trials before decisions need to be made.

The trial duration is likely to be in the order of 10 to 15 years because of the anticipated slow natural spread of the pathogen, and the complexities of the forest systems with which we are working. Therefore, trial design and set-up needs to be very carefully done to allow people other than the initial project team to continue the work if necessary. Although it may take many years to get definitive results on pathogen spread in the soil, and to be confident about the effectiveness or otherwise of phosphite barriers, preliminary data and useful information should be provided regularly. The anticipated trial duration to achieve definitive results will probably outstretch current funding options, but provided the trials are properly established and work is progressing well, there should be no problem in renewing funding when required. The most significant costs will be in the establishment phase of the trials, probably over a two year period. Thereafter, activity will decline to periodic monitoring of disease progress, with occasional flurries of activity for phosphite treatment re-application or plant/microbial community studies.

Extreme care must be taken to avoid compromising the barrier work by excessive studies or movement on site. However other projects could potentially tie in with the barrier project. If plots are well set up they could be a mine of information in future years. But the project team must keep reminding themselves of the key question — does a phosphite barrier slow the natural rate of spread of *P. agathidicida* through a site?

The nature of the barrier project means that it cannot be accurately costed until the investigation and delineation of potential sites is carried out. There will have to be a multi-step process with re-evaluation points throughout. Once a project team is selected and broad trial specifications are determined (based on the discussion in this report), the first requirement will be to scope, roughly map, and do preliminary soil sampling and testing of suitable trial sites. In turn this will

determine most of the other trial criteria, such as plot size, number of plots, potential assessments that should be made, and ultimately the cost of setting up trials. This part of the work will take considerable effort, probably many months. The time and cost to complete this task is unpredictable until the work is done. There will need to be a degree of flexibility in project planning and contracting that allows for changes to be made as required, so that correct decisions can be made without compromising the project. This flexibility should continue throughout the project, with key re-evaluation or stop/go points written in. It will require a partnership and trusting interactive relationship between researchers and funders to allow flexibility, especially over the first year or two of the programme. This won't necessarily fit the model of 'prospect, tender, contract and do' that is currently in vogue. An oversight group, comprising the Kauri Dieback P&I team, or other advisors, might be a good way of managing this.

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1 INTRODUCTION/BACKGROUND

Kauri dieback, caused by *Phytophthora agathidicida*, is causing significant losses of kauri trees in Auckland and Northland. Long distance spread is probably caused mostly by human factors such as soil movements on footwear, machinery and planting material, perhaps enhanced by roaming animals such as pigs. In infected stands there is also a slow natural spread of the pathogen either in the soil or from root-to-root, resulting in a steady increase in the size of infected patches in the forest.

Few tools are available for the control of *Phytophthora* diseases, particularly in natural ecosystems and forests. Treatment with phosphite is probably the most promising treatment available, with this chemical being widely used for *Phytophthora* control in horticultural crops such as avocados, strawberries and apples. There has also been a considerable amount of research carried out on using phosphite for management of *Phytophthora* in native ecosystems, with the majority of this work carried out in Australia. There is a significant amount of literature on the treatment of various native species and plant communities by methods including trunk injection, knapsack spraying, aerial spraying from aircraft and trunk sprays (e.g. Crane and Shearer 2014; Pilbeam et al. 2000; Shearer and Fairman 2007). Numerous authors have published studies on phosphite uptake, residual activity time, phytotoxicity, host responses, and species variability. In New Zealand kauri forests, phosphite has been shown to suppress the extension of *Phytophthora* lesions in kauri seedlings and ricker trees, and to provide protection against root infection (Horner and Hough 2013; Horner et al. 2015). Once rates and treatment regimens are refined, it is expected that phosphite will provide a useful tool for the management of diseased kauri trees and stands.

The concept of using phosphite as a barrier to reduce spread is taking individual tree treatment a step further. In theory, treating a band of vegetation or potential host species ahead of the disease front will not only protect those treated trees from infection, but also reduce the rate of pathogen spread through the site by directly reducing the root-to-root spread. Such treatment could potentially be used in kauri forests to slow the rate of spread of *P. agathidicida*. A first step is to demonstrate that the barrier treatment could be beneficial, but this ‘proof of concept’ will be very challenging. The purpose of this report is to (i) review past work on phosphite barriers, (ii) identify some of the factors that should be considered in research and proof of concept trials in kauri forest, (iii) to recommend a logical progression of required work, and (iv) to assess the feasibility of such work.

A brief review of work related to phosphite barriers overseas and in New Zealand, and commercial phosphite barrier treatment operations in Australia, is provided in the Appendix.

2 REQUIREMENTS FOR PHOSPHITE BARRIER TRIALS IN KAURI FORESTS

The challenge in New Zealand. Proof of concept and difficulties in obtaining this.

In discussions with fellow researchers in New Zealand, Australia and the USA there is a general belief that phosphite barriers should reduce natural spread rates of *Phytophthora*, and that the technique has potential for use in some kauri forests with dieback problems. But even though the concept of phosphite barriers is sound, proof of concept in kauri forests will be difficult. The real challenge is to carry out unbiased experiments in a way that can prove the effectiveness (or otherwise) of the phosphite barrier system. Many factors in the New Zealand bush will make this difficult, long-term and costly. The following are a series of requirements and points to consider regarding trials to test the efficacy of phosphite barriers to retard the spread of kauri dieback. In part they relate to points identified in the draft Phosphite Barrier Discussion Document prepared for MPI in early June 2016, and further discussed at the Phosphite Barrier Workshop held at the Mt Albert Research Centre on 17 June 2016 (circulated previously).

2.1 Trial site requirements

Selection of appropriate trial sites is essential to facilitate practical and useful trials. The trials will be in place for many years, so it is very important that good decisions are made at the start. Ideally, sites should be relatively flat with even aged stands, with a high stocking of kauri, good demarcation of diseased and healthy areas, sufficient diseased and healthy areas to allow for the replication of treatment plots, and relative ease of access. In reality, this ideal is very unlikely to be met and compromises will need to be made. Nevertheless, certain parameters *must* be met for a site to be considered for the trial. These include:

- **confirmed *P. agathidicida* presence**
- **contiguous 'clean' forest**, so that the rate of spread into this area can be assessed
- **sufficient area** to allow at least one treated and one untreated control plot at each site
- **moderate to high stocking of kauri**
- **permission** from forest owners or key stakeholders for long-term work on the site

None of these above five parameters can be compromised.

A number of other factors should also be considered in site selection. Whilst these factors could potentially be compromised, the project team should at least consider these points in selecting trial sites.

Slope: While relatively flat sites would be preferred for the trials, it is unlikely that such sites are available. Therefore, the slope of the site will need to be considered when laying out plots. Uphill slopes would be preferred for measuring natural spread from diseased to healthy areas (i.e. the spread uphill from diseased to healthy areas), as spread will be less influenced by rapid run-off events that could impact downhill spread. Uphill spread might also have most relevance to the eventual use of barriers in reducing spread from one catchment to the next. However, the measurement of downhill spread could also be carried out, where possible, as a comparison potentially providing useful information.

Vector control: This will be very important, as high pressure from potential vectors, such as pigs and humans, could completely compromise trial integrity by spreading inoculum and 'leapfrogging' barriers. The canopy in dieback areas may be thinner, resulting in wetter soils and, as a result, be more attractive for pig wallowing which could have a major impact on spread. Therefore, pig culling may be necessary in some areas. Where required, appropriate signage should be used to minimise human entry. Fencing some sites could be considered, but will probably not be practical in most areas. Effective 'island' stands, such as isolated small stands or pockets of fenced bush on farmland, may be useful as sites may be less impacted by mammalian vectors.

Kauri size class: In mature kauri forests, the large trees and sporadic spacing would make plot selection difficult. The very wide-spreading root systems would mean that plot sizes would have to be very large to minimise interaction between plots. In ricker and medium-sized kauri stands there is usually a higher proportion of kauri to other species, and the root systems are not as wide spreading. In these stands the plot size could potentially be smaller than for mature stands, allowing better replication and more manageable plots.

Vegetation type and geographical area: While it might be desirable to select sites in certain geographical areas or vegetation types, it is unlikely that this will be a major consideration if other key parameters are met. In meeting the other parameters, it is likely that trials will cover a range of geographical areas and vegetation types, which will have to be considered in trial design and analysis. Suitable replication and balance in design should be able to cope with different geographical and vegetation factors.

Site access: Very public sites should be avoided because public interference, such as walking through sites or disturbing tags or markers, could compromise trial integrity. However, from a researcher's point of view, relatively easy access is desirable, especially if equipment, for example, is required on site for either treatment or assessment. Remote access by foot will also impact on the time required for treatment and assessment, however, if other trial parameters are met, access issues will be dealt with as required. Apart from treatment application and the possible need for spray equipment (see below), heavy equipment is unlikely to be required. There is always the option of equipment/materials dump by helicopter.

Avoidance of other studies: Care will need to be taken when selecting trial sites to ensure that other studies will not be compromised. We *expect* phosphite to have an effect, so the barrier trial should avoid sites where other researchers might, for example, have plots monitoring dieback effects or looking at seedling recruitment.

2.2 Potential trial sites

As a starting point in site selection, all Kauri-dieback-confirmed sites should be considered. To do this, the project team should have access to the complete distribution records to date. This is not currently available. Among the pool of researchers at the 17 June workshop, different people had knowledge of different areas of forest. At this stage few people have a good overview of *P. agathidicida*-positive sites.

A number of potential trial sites have been suggested but it will take an in-depth discussion between researchers and people familiar with the sites to determine which might be suitable for the trial and which could best meet the main criteria. Once this initial screening has been done, appropriate permissions and preliminary site visits would be required to finalise site suitability and subsequent trial design.

Suggested sites include the Waitakere Ranges (multiple site possibilities), Coromandel Peninsula, Great Barrier Island, Glenbervie, Raetea, Omahuta, among other possibilities. Results of recent sampling in Northland, suggest that many small sites in this area may be useful for barrier trials. Old forestry plantation blocks could potentially make good trial sites because trees would be located in even aged stands and evenly spaced. There would be both advantages and disadvantages at such sites. All potential sites would need to be assessed to see if they meet the key trial criteria.

Comments on a few known sites:

Omahuta This was suggested as a potentially useful site as the infected site has a narrow band of kauri trees planted in a line, with uninfected trees at each end. This could have been an excellent site for treating one end and leaving the other as an untreated control. However, a recent inspection of the site has determined that at the northern end of the stand the infection has spread significantly from what was mapped 4 years ago, leaving only one tree not showing symptoms. This spread of infection means this plot is not suitable for the barrier trial.

Raetea Forest This is another plantation site with definite potential for barrier studies. There are many infected trees in the site and from a distance the disease appears to be occurring in patches, with some apparently healthy areas. In order to use this site, detailed mapping and sampling will need to be carried out to determine just how widespread the infection is, and if the 'healthy' areas are in fact pathogen free. The site is also highly disturbed by pigs which could be a problem.

Albany Reserve and Okura Reserve Both of these sites have relatively small confirmed patches of kauri dieback in regenerating stands, with potential for barrier trials. One drawback is that the sites are both very public, with potential for interference (despite track closure).

Huia Dam Near the ridge in the area above the current phosphite trials, there appears to be a wave of infection gradually spreading through the stand of regenerating kauri. This site has good potential for a barrier trial. Access to the site is relatively easy, yet about 200 m away from a track, so not too public.

Cascades There are many stands in the Cascades region that are confirmed with kauri dieback, particularly near the Upper and Lower Kauri Tracks. Some could potentially be useful for phosphite barrier studies. Much of the area is very steep which may make studies more difficult, but there could still be some suitable areas.

Waitakere Ranges There are multiple infected sites in the Waitakere Ranges. It is highly likely that some of these will be suitable for barrier studies. Recent/current mapping studies carried out by Auckland Council will be helpful in this.

Waipoua and Trounson Forests Both Waipoua and Trounson have significant kauri dieback infections, but these forests would probably not be suitable for the initial barrier trial work. The predominantly large trees would make selecting suitable plots difficult, and plots would need to be very large. This does not mean that these forests would not be suitable for treatment using phosphite barriers as a tool to protect areas or contain infections in the future, should the local guardians choose to do so.

Site Variability.

Ideally, sufficient replicates would be located within the same forest. This would minimise problems with vegetation and other site differences that may confound disease development or progression. However, it is highly unlikely that this ideal will be met. A likely scenario is that one or two replicates at most will be possible within any forest or stand, and replication will have to be across different stands, perhaps even different regions. This is not necessarily a problem in terms of interpretation of results provided the design is well balanced, essentially treating different forests as 'blocks' in an experimental design sense. In fact, such a geographical spread could be seen as an advantage, giving the opportunity for learning how kauri dieback progression and responses to phosphite may vary under different environmental and other scenarios. Provided that sufficient site parameters are assessed at the start of the trial (see Section 2.8), these could be treated as co-factors in subsequent analyses, potentially revealing information about kauri dieback ecology. The key question is whether or not there is differential spread of kauri dieback in treated versus untreated plots. The forest the work is done in is incidental to this question, and diversity of sites could be beneficial in the long run.

2.3 Stakeholder Consents / Permits

In the selection of sites, land ownership and interests must be considered both in the short- and long-term. These trials will be expensive to establish and will take many years to complete, so continued access is vital. Owner/stakeholder involvement, understanding and acceptance of the programme is important. As part of this, consultation with landowners, manawhenua and other local groups will be important from the early planning stages. Where land is under Department of Conservation (DOC) or Council jurisdiction appropriate approvals will be required, which will include consultation with local iwi groups.

Gaining consent from landowners, managers or manawhenua should not be the end of the consultation with these groups. Throughout the trial process there should be regular interactions with key stakeholders, keeping them abreast of trial progress, developments and practical outcomes, and giving them the opportunity for comment. Where practical, representatives of owners or manawhenua could be invited to participate in some aspects of the trial work, while bearing in mind that the sensitivity of the trials plots and potential for confounding influences will require strict limits on site access and activity.

Public and private concerns about treatments in forests needs to be considered from the start. It is important that public and private questions (e.g. about environmental effects, toxicity etc.) are carefully considered and addressed as part of the trials. Issues arising at this stage are also likely to be raised in the future if, and when, broader management treatments are proposed, so it is important that such questions are addressed as part of the trials.

Another factor that should be considered or clarified by the Kauri Dieback Programme (KDP) in advance of trials is whether or not approvals are required from ACVM or other regulatory authorities for use of phosphite in the planned manner. This is a very grey area at present.

2.4 Demarcation of infection fronts[1]

The main aim of the barrier trial work will be to determine if treatment slows pathogen spread through a site. This will rely on assessing the change in pathogen distribution over time. The best way of doing this will be to monitor the differential spread of the infection front in treated and untreated plots. To do this in a timely way, it is essential to have accurate demarcation of the disease front at the start of the trial and an ability to measure it accurately to determine differential spread rates. The aim should be to have the infection boundary marked to within approximately 1 m. More accurate infection front delineation will potentially allow for smaller plot size and more rapid discrimination in rate of spread among different treatments (see further discussion at the end of this section).

Accurate infection boundary discrimination will be difficult in kauri forests. Unlike studies in Australia, where numerous herbaceous or shrubby indicator species can help delineate disease margins to within a metre or so (Shearer et al. 2004), there are no known indicator species in kauri forests other than kauri itself. The main problem is that kauri trees may take many years (perhaps decades) from initial infection until when they show above-ground symptoms, such as basal trunk lesions or canopy thinning, especially in larger trees. By this time the infection is likely to have spread many metres beyond the tree. Therefore, there is a need to delineate the infection front in some other way.

Soil baiting. At present soil baiting is the most effective and accurate technique for determining the presence of *P. agathidicida* in the soil, and is probably the easiest option for monitoring pathogen progression. Although false negatives are possible, the impact of these can be minimised by an appropriate sampling regime. Fine-tuning of the soil baiting system is possible, but recent experiences suggest that the system is accurate and repeatable, within the limits of natural biological distribution and variability. A structured sampling regime will be required that, in a series of transects or grid samples over time, gradually increases focus on the infection front. This will help minimise the amount and cost of sampling required, without compromising accuracy.

DNA detection techniques. At present DNA-based detection techniques are not considered accurate enough for the testing required for the infection front delineation. There is also no confidence that they'll be able to adequately screen the large composite samples of soil required to detect sporadically distributed presence. However, as technology improves such systems are likely to become available during the course of the trial, potentially improving the sensitivity and cost of the testing. In the meantime, assuming soil baiting systems are used initially, it would be worthwhile storing a portion of all samples for later analysis if technology improves.

Natural indicator species. Is there a sensitive indicator species that could demonstrate the presence of *P. agathidicida* in the soil? Although this is a possibility (see discussion on hosts below) no such species are currently known. Even if suitable species are found, they will be of little use unless they have broad natural distribution in kauri forest. Is there a difference in community structure in infected and uninfected kauri forest, and is this difference sufficient and expressed early enough to be of use in delineating a disease front? Little work has been done in this area, and long-term studies would be required to determine such factors.

[1] In this document, the term 'infection front' refers to the actual distribution of *P. agathidicida* on the roots or in the soil. 'Disease front' is a more general term, usually referring to the expression of symptoms (e.g. canopy thinning, dieback or basal trunk lesions). The disease front will generally lag some way behind the infection front, in both a spatial and temporal sense.

Planted indicator species. If an indicator species is found, these could be planted across the disease front in trial plots so the pathogen progression can be mapped readily. If such a plant could be found it would require an ability to grow in the challenging environment on the forest floor in kauri forest, and be highly sensitive to *P. agathidicida*. It would need to be non-invasive (i.e. not weedy) and would need to be removed at the end of the trial. Perhaps kauri seedlings, either naturally occurring or planted, could be an appropriate sentinel species.

Indicators on kauri trees. Biological indicators, such as lichen growth on kauri trunks, may indicate diseased trees. Healthy, rapidly-growing trees shed bark regularly, so build-up of lichen or other epiphytes on kauri trunks may reflect slower tree growth. But would this be timely enough to be a useful indicator of the disease front?

Accuracy of front demarcation. It will be difficult to discriminate infection boundaries to within the 1 m target suggested above, regardless of the techniques used. This sort of finely-tuned delineation has not yet been attempted with kauri dieback. To determine what is realistically achievable in margin delineation, it is suggested that a detailed pilot study is done on a dieback site, preferably in one of the prospective barrier trial sites. This could give an indication of what is reliable and repeatable, and what sensitivity can be realistically achieved. This in turn will help predict what sort of time period may be required to discriminate infection boundary advance between phosphite-treated and untreated plots (i.e. poorer infection boundary discrimination will mean that a longer trial duration will be required to confidently gauge a treatment effect).

Relationship between infection front and symptom front. In addition to delineating and mapping the progression of *P. agathidicida* through soil over time (i.e. the infection front), the expression of symptoms in kauri trees should also be mapped and followed. This would be a much quicker and cheaper assessment, although it would not necessarily reflect the underground spread of the pathogen. By studying both the infection front in the soil and the expression of symptoms in trees, it may be possible to determine a consistent spatial relationship, perhaps related to site factors or tree size class. This would give insight into the lag time between pathogen presence and tree symptom development/decline, plus indicate what sort of buffer areas should be considered beyond trees with symptoms. Such information would be particularly useful to forest owners/managers interested in managing kauri dieback.

2.5 Plot size

The required plot size for phosphite-treated and untreated areas will vary depending on multiple site factors. In Western Australian (WA) trials by Shearer et al (2004), plots were 10 x 15 m, with replicate plots lined out along a disease front. However, their trial took place in a shrubland/small tree forest type. In kauri forests, with larger trees and potentially wide spreading root systems, plots may need to be larger to avoid too much interference between adjacent treated and non-treated plots.

Factors to be considered when determining plot size may vary with each site, and include tree size, topography, vegetation type and drainage. The potential rate of spread should also be considered when determining plot size. Expected rate of autonomous spread (*P. agathidicida* growth) is up to 4 mm/day (from twig inoculation assays, Horner & Hough 2014), i.e. approximately 1 to 1.5 m per year. That is assuming just pathogen growth, with no assistance from localised potential vectors, such as invertebrates and rain-splash, for example. With these local vectors added, we might expect spread between 1 and approximately 4 m per year. Note that this estimate is for a flat site or uphill slope. Because of the potential for mass water

movement carrying soil and spores, downhill spread could be substantially greater and very difficult to predict.

Other points to consider:

- In general, plots should be wider than they are deep to allow for lateral interference between plots, with measurements taken only towards the centre of plots.
- Root grafting, especially with larger kauri trees, could potentially spread phosphite from treated to untreated plots.
- Where large trees are present, roots could span plots causing problems with treatment integrity. If plots are too small there could be untreated tree roots crossing treated plots, potentially breaching any phosphite barriers. Therefore, treated plots must be big enough to avoid this problem. In general, sites with larger trees should have larger plot size, plus possibly a wider treated barrier.
- Root distribution/spread of kauri and other trees is not well understood and difficult to investigate. Some basic research on typical root spread in advance of setting up plots would be useful to at least allow better prediction of plot size.
- The plot depth should be greater than the treated band depth (i.e. the barrier), to allow monitoring of pathogen spread beyond the barrier. In the long-term, this could be an effective way of assessing barrier effectiveness.
- Plots should be laid out perpendicular to the disease front, with the initial edge of the plot on the disease front.
- Ideally, plots should be perpendicular to the slope to account for water runoff, but the need to be perpendicular to the infection front is more important. As a compromise, provided bulk water flow is from the non-infected to the infected area, exact perpendicularity to the slope is not essential.
- Plots should generally be laid out on the upslope side of an infected area (see earlier discussion), although some downhill plots would be useful for comparison.
- For reasonable statistical analyses, at least five replicate plots of each treatment would be required. However, given the likelihood that any trials will be spread over a range of sites, and the high variability in multiple site parameters at those sites, a much higher number of replicates is recommended (10 to 12)

At the workshop on 17 June, there was considerable discussion about plot size, but little agreement. General consensus was that the 15 x 10 m plots used by Shearer et al (2004) would not be large enough, but estimates for the kauri work ranged from 15 x 20 m to 100 x 100 m. There needs to be a balance between what is desirable and what is practical and manageable. In ricker, or moderate sized tree stands, plots of approximately 20 m deep x 30 m wide should be adequate.

Related to plot size, the depth of the treated area (barrier) needs to be considered. In Australia, 15–20 m is recommended and has been found effective even with widely spreading root systems (Tuffnell personal communication). For kauri forest a 15–20 m barrier zone is probably a good starting point for the trials, particularly if trials are done in regenerating ricker stands. If treatment is to be adopted on a large scale in the future, it needs to be realistic and practical.

2.6 Target plants for treatment

The principle of a phosphite barrier is to effectively provide a host-free barrier to prevent pathogen spread. By treating otherwise susceptible trees, the phosphite effectively turns them into non-hosts. A phosphite barrier can only be effective if the majority (or all?) of the host species in the barrier zone are treated. With kauri dieback this is problematic, as at this stage we do not know the host range of *P. agathidicida*. Kauri is the only known susceptible host tree in the forest, or at least the only tree proven to show significant symptoms and death. But anecdotal and some experimental evidence shows that other species could potentially harbour of even proliferate the pathogen. For example, in the trials refining baiting systems for *P. agathidicida*, leaves of numerous bait species (including a number of native species) were readily colonised by zoospores, with subsequent development of lesions from which *P. agathidicida* could be isolated (Bellgard, Horner, Dick *unpublished*). In glasshouse inoculation studies, Bellgard demonstrated that a number of native species were colonised by *P. agathidicida* when grown in infested potting mix. Therefore, for the purposes of the barrier trials we should not assume that kauri is the only host, and other species could harbour or proliferate the pathogen without necessarily showing obvious disease symptoms.

This leaves a conundrum regarding what to treat in the barrier zone, with three main possibilities:

1. Treat just kauri
2. Treat all vegetation
3. Treat kauri plus selected vegetation based on knowledge of potential hosts.

The third option is not realistic at this stage as knowledge of potential hosts is not yet available, so we cannot selectively treat with any confidence. This leaves options one and two, with untreated controls for comparison, as potential treatment options.

There will be less potential environmental impact if only kauri are treated, and the work (both experimentally and in any future roll-out of the treatment) will be considerably less complex and cheaper. This would be the preferred option and one that should be tested. But the counter argument is that it may not be effective if significant numbers of alternative hosts could be maintaining or proliferating the pathogen, so we also need to include the ‘all vegetation’ treatment option.

To improve the potential barrier effectiveness, it makes sense to also treat the zone of infected trees immediately behind the infection front. Many of these trees are likely to have root systems extending into the barrier zone and would be an ongoing source of inoculum if left untreated.

2.7 Treatment regimens and application methods

For trees, the most effective phosphite application system is via trunk injection. Thus, for trees large enough to inject (i.e. trunk approximately 8 cm diameter), injections should be the main application method. For smaller trees, shrubs and seedlings, foliar spray is probably the most practical phosphite application method. This reflects the regime used in phosphite barrier treatments applied on a large scale in WA.

The scale of the trial, and the predicted difficulty in finding suitable sized sites meeting the key trial criteria, means that the number of different treatments must be kept to a minimum. Thus there is a need to make a best guess estimate of spray or injection

concentrations/regimes and leave the investigation of alternative rates and regimes for other trials. Some preliminary work could be done to inform decisions on rates.

There is an argument for keeping rates as high as possible as this will potentially increase the effectiveness and longevity of any treatment, and aid in the initial proof of concept required in this work. But this must be balanced against the potential for phytotoxicity (e.g. leaf burning and leaf drop) which may not be acceptable to some groups.

Trunk injection: There is already some good data available from trials with kauri to show that phosphite injection is effective at controlling *P. agathidicida* (Horner et al 2015). However the trials described in this work only investigated phosphite concentrations of 20% and 7.5%, injecting 20 ml of solution every 20 cm of trunk circumference. On some sites phytotoxicity symptoms were noted with both of these concentrations, especially the 20% rate. Current trials, established in March 2016, are investigating lower concentrations (4% phosphite) and more widely spaced injection points (40 and 80 cm), effectively applying much lower amounts of phosphite. By the time the barrier trials are established, there should be some data available on whether or not these lower rates avoid phytotoxicity, and yet are still able to control *P. agathidicida* growth. Thus appropriate injection rates for kauri can be determined before trials start.

No information is available for other tree species that might be injected in some trial sites (e.g. rimu, tanekaha, totara, and taraire). Responses and sensitivity among different species are likely to be highly variable. In WA, a default concentration of 5% phosphite is used for trunk injection across a wide range of tree species where there is no detailed knowledge of effective rates or phytosensitivity. It is recommend that the 5% concentration is applied for injections in the proposed barrier trials, unless information generated in the meantime contradicts this.

Foliar spray: Foliar spray application has not been investigated in New Zealand native forests, so application rates that are effective and safe are not yet determined. Some insight can be gained from the work of Scott et al (2016) who investigated the phytotoxic responses following phosphite spray application onto container-grown plants of a number of NZ native species. They found only minor phytotoxic effects across a range of taxa and phosphite concentrations. Even where there were phytotoxic effects, the plants subsequently recovered and re-grew. In broad applications in the WA bush, a phosphite concentration of 0.5% was used for spray applications of the understory and shrub layer. This concentration, sprayed to runoff/drip, is probably an appropriate rate for trial applications in kauri forest. While phosphite response will vary among plant species, for practical purposes a single application rate must be selected for forest under-storey applications.

Phosphite uptake through leaves is enhanced by the addition of wetting agents or surfactants. Care must be taken to avoid surfactant which may inhibit phosphite activity. The appropriate surfactants and rates for use in the NZ bush are yet to be determined. In advance of the barrier trials, small-scale pilot studies should be carried out on shrub-layer plants in the NZ bush to determine appropriate spray rates and surfactants. A pilot study to spray kauri seedlings will also be needed to understand, for example, potential phytotoxicity issues and surfactant requirements for leaf uptake.

Phosphite treatment is a control and not an eradicant. Even if *P. agathidicida* could be cleansed from a tree, the pathogen would still be present as spores in the soil and organic matter, and could recolonise trees once phosphite concentrations in tissue have dissipated. Therefore, repeated application of the barrier treatment will be required, probably every few years. In kauri trials, there has been only negligible re-activation of lesions four years after phosphite

application (Horner unpublished), suggesting that a 4 or 5 year interval between treatments may be acceptable. In WA, evidence suggests that injection treatments last from 3 to 5 years, and sprays from 1 to 3 years. As a compromise, commercial re-application of both injection and spray treatments is generally carried out every 3 to 4 years. A similar regime will probably be required in kauri forest trials, perhaps aiming to stretch the interval as long as possible, dependant on findings from current phosphite trials. By the time decisions need to be made about retreatment in the proposed barrier trials, substantially more information will be available from existing trials, allowing more informed decisions.

2.8 Measurements required

The main purpose of phosphite barrier trials is to determine the effectiveness of phosphite barriers at slowing the local spread of *P. agathidicida* in the forest. Thus, the key measurements will be those directly assessing pathogen spread and comparing differential spread in treated and untreated plots, particularly with respect to movement of the infection front. The other essential measurements relate to assessing impacts of phosphite on the treated plants and the overall ecology of treated areas. This will be particularly important if spray application of the understorey is carried out. Many other measurements and studies that may help elucidation of the ecology of kauri dieback could potentially be made as part of these trials, but these must not interfere with the key driver of the work.

The most important assessment throughout the trial will be the presence or absence of *P. agathidicida* at points beyond the originally mapped disease front. The sampling regime will be important and should be determined by the project team taking into account site factors etc., but periodic sampling along prescribed transects in treated and untreated plots would be a good way of doing this. Over a period of time this assessment alone should give sufficient information to answer the key question about whether or not phosphite barriers slow the spread of *P. agathidicida*. As discussed earlier, soil baiting is the best current technique for making these assessments, although the project team should remain open to the possibility of improved techniques becoming available during the course of the work.

However, mapping of the infection front boundary shouldn't be the only assessment made. There are many site parameters that should be investigated at the start of the trial, such as vegetation type, slope, aspect, drainage, and soil type. A number of these could have some influence on the outcomes of the trial and will need to be treated as co-factors, especially in the likely event that replicates in the trials will be spread out over a number of different sites.

Disease indicators others than *P. agathidicida* presence or absence in the soil could also be useful. At the very least, kauri dieback symptoms such as canopy thinning or basal trunk lesions should be regularly recorded. Periodic kauri feeder root health could also be a useful measurement, although sampling may cause a degree of interference in the trial that is undesirable. Root measures could perhaps be made in conjunction with soil sampling for presence/absence testing, although the root health results obtained in this instance may add little to the subsequent baiting data. 'Cultural Health Indicators' should also be considered where useful indicators can be identified, with consultation and involvement with local manawhenua.

Potential assessments to be made:

Initial assessments: Soil testing for *P. agathidicida* presence must be done, focusing on determining the boundary between 'infected' and 'clean' zones. All soil sample points should be marked in the field, to back up any GPS or other data collected. Precision is important. Clean and infected zones should be clearly marked on the map and in the bush (by tagging trees and using flagging tape).

Sites should be accurately mapped, noting slope, aspect and drainage patterns. Traditional methods, or perhaps Lidar, could be used for this. Tracks or other disturbances should also be marked.

All kauri (including saplings) should be mapped, along with other significant trees (>5 cm diameter?) with general notes on vegetation type and canopy structure. Soil type could be assessed, along with some basic measurements of, for example, nutrient status and pH levels at each site. Rainfall or general climatic data (if available) might be useful. Pig damage or signs, and indications of human interference should also be noted.

It is important that broad and robust baseline measurements are made, as this will potentially facilitate unforeseen studies in the future.

Post-treatment assessments: Toxicity symptoms in kauri or other species should be assessed, in particular looking for differences in treated and untreated zones. Special attention should be paid to lichens and mosses which might be particularly sensitive to phosphite sprays. Measurement of phosphite residues in soil or non-target species at intervals post-treatment may be useful, but appropriate techniques would need to be identified first.

Regular assessments: Yearly or perhaps biennial assessments should include kauri tree survival, symptoms (canopy and basal trunk), and soil sampling for infection front tracking (especially on transects). In the future, spectral imaging may be of use in determining tree health, but in the meantime visual observations of canopy and trunk lesion changes such as those made in the current phosphite efficacy trials are adequate.

Other potential assessments: Ecological plots could be established. These could be designed to look for changes as *P. agathidicida* progresses through plots, and to look for possible phosphite impacts. Useful information could be gained from assessment of community structure in phosphite-treated and untreated, infected and non-infected zones. Such assessments might include seedling succession and recruitment. Although these ecological assessments may not contribute directly to the goal of differential spread rates, the structure of the proposed trials are potentially a good opportunity for such studies.

Kauri tree growth could also be monitored (e.g. with growth bands), or growth could be assessed retrospectively using dendrochronological techniques. In the long term, such measures could give some insight into tree responses to infection by *P. agathidicida*, with or without phosphite application.

In the future, measures of microbial activity over time could be made using techniques such as quantifying ecological DNA, looking for soil functional changes in response to phosphite application. Again, while not directly contributing to determining barrier treatment efficacy, such measures could be a useful addition to the work.

Whatever assessments are made, there must be very strict and rigidly enforced rules around site access and disturbance. Lack of care could potentially compromise the trial by spreading infected soil from 'infected' to 'clean' areas, confounding results. Rules might include:

- Restrict assessments to dry periods.
- Always work from clean to contaminated direction.
- Minimise risk by limiting sampling to once per year or more.
- A raised plank or boardwalk system could possibly be employed to minimise vectoring soil through sites, but this may not be practical in the dense bush and rough terrain typical of most kauri forest.

Although the barrier trials might be an opportunity to make all sorts of ecological measurements to help elucidate ecological questions regarding kauri dieback, such studies should probably be focused elsewhere, and the barrier sites only utilised where unique opportunities exist. Focus must remain on phosphite impacts (both positive and negative) on tree health, forest ecology and disease spread.

2.9 Ecological impacts of treatments

Phosphite application could potentially have some ecological impacts in treated stands, particularly if spray application of the understorey is undertaken. It is important that any trials looking at barrier treatments take into account possible ecological impacts (positive and negative) and try to describe and quantify these.

Possible impacts might include direct phytotoxicity, changes in phosphorous status of soil, composition of mycorrhiza, soil microflora, lichen and bryophyte composition, or species composition of regenerating seedlings. All of these possibilities should be monitored to determine if they occur and to gauge their importance. To avoid unforeseen impacts and to address any public concerns about treatment, such issues would need to be addressed in trials before any future wider roll-out of barrier treatment. With such information available, informed decisions can be made about whether to treat, and the consequences (positive and negative) of doing so. Effects should be kept in perspective given that treatments are applied only once every 3 to 5 years, so off-target impacts may be ephemeral. There will need to be a balance between the ecological impacts of phosphite treatment and the ecological impact of unrestricted kauri dieback spread.

The design of the barrier trial, essentially a replicated block design, should allow for direct comparison of various ecological components between treated and adjacent untreated plots. Impacts of treatments on species composition, whether at the higher plant, bryophyte or microbial level, could be made at intervals or at the end of the trial, directly comparing treated and untreated plots. To complement any subsequent measurements, a baseline assessment of species composition and health carried out at start of trial would also be useful, although not essential.

While the phosphite ecological response assessments should be made as part of the barrier work, separate and dedicated trials should also be carried out to minimise disturbance of the barrier plots. Such trials don't necessarily have to be carried out in kauri dieback-affected forests.

The mycorrhizal status of phosphite-treated and -untreated plots is one area where there should be some detailed studies. While this topic also requires a dedicated and detailed investigation separate from the current study, the barrier plots potentially offer an excellent opportunity for assessing impacts in the forest, on both kauri and other plant species.

Phytotoxicity symptoms, in particular leaf scorching or yellowing should be assessed and quantified on kauri and all other species treated. Preliminary work by Scott et al. (2016) is useful, and could be expanded on. This could be accompanied by phosphite residue testing to determine retention of the chemical within plants. Soil assessments (if possible), could give an indication how much is reaching the soil, although concentrations might be expected to be below the detection threshold.

Concerns have been expressed that the amount of phosphorous added in the phosphite application could contribute to the phosphorous concentration in the soil, perhaps influencing a vegetation adapted to low P levels. However this seems unlikely. A crude calculation, assuming a typical soil has a P concentration of 200 ppm, with a soil density of 1.6 g/cm^3, gives 640 kg of P per ha in the top 20 cm. Assuming an application of 300 ml of 5% phosphite into a tree with 1 m trunk diameter and 10 m canopy diameter, this would add the equivalent of about 0.7 kg of P per ha. This is just over 0.1% of the total P in the soil. Considering that most of the P in the phosphite would be retained within the plant and later bound into organic matter for a long period of time, this is not seen as a problem.

There could be many potential opportunities in this trial to carry out ecological measurements of kauri dieback impacts. This knowledge is currently lacking because little work has been funded to date. But this work also needs to be focussed elsewhere to minimise disturbance to barrier plots, where the main purpose should be assessment of differences in disease advance in treated and untreated plots, with checks on potential ecological impacts.

2.10 Skill requirements for project team

The project team composition will depend entirely on the scope of the work required.

In its simplest form, addressing solely the question of whether phosphite barriers slow the rate of *P. agathidicida* spread in the forest, the work could be carried out by a competent pathologist with reasonable knowledge of the NZ bush.

By including ecological measurements and impacts, an ecologist and botanist (including someone with expertise in bryophytes and lichens) would also be essential. Input from a statistician/biometrician will be useful during the design and analysis phases.

People will skills in detailed mapping (potentially using UAV technology) would be useful, particularly in the initial stages.

Services such as soil chemical analyses and residue testing could be separately commissioned, and such expertise would not have to be part of the project team.

2.11 Associated research required (either before or simultaneous)

While the proposed phosphite barrier trials could be done independently, there are a number of related aspects of work that could or should be done in advance or in association with the proposed trials. These are briefly summarised below.

Host range There is still little direct knowledge of which plant species present in kauri forest could harbour or help proliferate and spread *P. agathidicida*. This lack of knowledge impacts the trials in that target species for treatment are not known. By default, everything needs to be treated (as one option), or pathogen spread could continue unchecked.

Indicator species Related to the host range work, is there a species that could be a useful indicator of *P. agathidicida* in the soil? Such a species (if it exists) could be usefully employed in the barrier trial as a tool for visually following subterranean pathogen spread.

Root spread 'How far do kauri tree roots extend in the forest?' This is currently unknown, but has a substantial bearing on the barrier trial. In particular, we need to ensure that the barrier dimension spans significantly more than the root zone of individual trees, or the barrier could be readily bridged. Similarly, plot width will need to be sufficient to allow for sideways interference from widely spreading roots from trees in adjacent plots. Some preliminary work on assessing kauri root spread could help inform subsequent barrier trial design and determination of barrier plot dimensions required.

Mycorrhizae Although there have been some studies on kauri mycorrhizae, nothing is known about the impact of phosphite on these associations. The barrier trials could be a useful place to assess any impacts on mycorrhizae in the long term, but we should avoid unnecessary disturbance of the soil in trial plots. Ideally, studies of mycorrhiza, and in particular changes that may occur in response to phosphite treatment, should be carried out in separate trials.

Phosphite spray application rates and surfactants Appropriate phosphite spray application rates for plants in the New Zealand bush are not known. While the easy option would be to default to the 0.5% spray concentration used in the WA treatments, there is an opportunity to do a small scale pilot study on shrub-layer plants in the NZ bush to determine appropriate spray rates in advance of widespread application in the barrier trials. This could help identify species that might be particularly sensitive. Apart from the work of Scott et al 2016, little is known about potential phytotoxic effects resulting from phosphite sprays on NZ native trees and shrubs.

Phosphite uptake through leaves is enhanced by the addition of wetting agents or surfactants. The appropriate surfactants and rates for use in the NZ bush are yet to be determined. In advance of establishing barrier trials, the alternatives should be investigated. A pilot study to spray kauri seedlings is required in order to identify potential phytotoxicity issues and surfactant requirements for leaf uptake. This work would not necessarily have to be carried out in kauri dieback-infested stands.

Non-target phosphite effects on plants, microbes and communities Within the barrier trials there will be plenty of opportunities to assess potential impacts of phosphite treatment on plants and microbial ecology. However, because of the potential disruptive nature of such investigations, these studies should be focussed on sites outside the main barrier trials.

Impacts of kauri dieback on plant communities The long-term nature of the proposed barrier trials will allow good opportunities for following changes in plant ecology as kauri dieback advances through sites. However, such work should be focussed at other sites.

Kauri root growth When are the peak flushes of kauri root growth? Does this have any impact on timing of phosphite applications?

Host defence response to phosphite Are there chemical responses within kauri that trigger the host defence mechanisms (e.g. changes in the Jasmonic acid pathway) and can we measure them? This could give an indication of when re-application treatments is required. Current work in the Healthy Trees Healthy Futures programme may address this.

2.12 Trial duration

The testing of phosphite barrier efficacy is likely to be a very long term study, perhaps up to 15 years to get definitive results. This is because of the anticipated slow natural spread of the pathogen, and the complexities of the forest systems with which we are working. This needs to be considered in trial design and set-up, so the work can continue under the direction of others not necessarily involved in trial establishment.

While definitive results might take many years to obtain, one might expect preliminary yet useful results on pathogen spread and barrier effectiveness within 4–5 years. Some related studies on factors such as phytotoxicity, impacts on other species, or changes in mycorrhizae (if they occur) might provide useful results even sooner. There could even be useful information coming from the very early reconnaissance and mapping stages, where relationships between trees with symptoms and subterranean spread of the pathogen could be determined.

While the anticipated trial duration might outlast current funding options, if the trials are well set up and providing useful information, it should not be difficult to get sufficient funding to continue barrier maintenance and monitoring in the future. Conversely, stop/go points should be put into the trial parameters, so that if for unforeseen reasons the trials are not yielding (or not predicted to yield) useful results, they can be terminated.

2.13 Costs

The cost will depend, in large part, on the scale of the trials and how many sites are required. This will depend on the suitability of the respective sites and how many replicates can be set up at each site. Ideally, the trial would be set up on one or two sites to test the barrier concept. But given the nature of kauri dieback, the complexities of kauri forests and the likely plot sizes required, it is unlikely that more than one replicate will be possible at each site, meaning that at least six to ten sites might be required to get meaningful and statistically valid results.

Because of the nature of the work, the project cannot be costed until the investigation and delineation of potential sites is carried out. This exercise would include the identification of potential sites, stakeholder consent, mapping and measurement of sites, and a significant level of soil sampling, allowing determination of the likely scale of the trial. There are probably many trial sites that could potentially be used for the barrier trials, but at this time a list of such sites is not available to the science community likely to be involved in trials. A full list of all infected sites should be made available to the project team so that appropriate sites can be selected. As a starting point, most kauri dieback sites could be seen as potential candidates for the barrier work, with a strict process of elimination imposed based on the key site criteria outlined in this report.

Initial set-up costs will be high. But once established, monitoring should be kept to a minimum, with measurements such as infection front mapping perhaps yearly or biennially. Assessments such as microbial or plant community effects might only be done once every 5 years or so. This will have the dual benefit of minimising disturbance and confounding impacts on the trial, while also keeping costs down.

2.14 Risks and barriers to success

There are a number of potential risks or barriers to success, but most of these should be able to be mitigated by careful planning.

- **Inability to find suitable trial sites**, including appropriate permissions. This seems unlikely given the large number of sites recorded with *P. agathidicida* presence, although meeting the strict trial requirements could be challenging.
- **Changing ownership of trial sites**. While this cannot be avoided, good communication should ensure new proprietors are supportive.
- **Changing attitude of owners**. A change of heart by landowners or guardians mid-way through the trial could jeopardise results. Owners could lose interest, or for some reason cancel rights of access. A real possibility, and one that occurs commonly in treatment trials in various situations, is that if early results indicate that treatments are successful, land-owners might want to treat all trees in the site, including untreated controls. Owners' rights should always be respected. Having trials spread over a number of sites with different ownership should partially mitigate any such problems.
- **Cost and ongoing funding security**. This work will be expensive and very long term. Funding security is important. There is potential to use KDP funding to help lever other funding (e.g. MBIE) for some aspects but this can't be guaranteed or relied upon.
- **Lack of knowledge of *P. agathidicida* host range** (including tolerant and susceptible hosts). If ignored and left untreated, alternate hosts could potentially compromise the work. This can be mitigated by having a treatment that includes all potential hosts.
- **Lack of knowledge of natural rate of spread.** This problem can be alleviated provided plots are large enough to cope with a range of potential spread rates.
- **Lack of knowledge of phosphite application and timing**. We should have more information regarding this by the time trials start, but there will still be unknowns. Pilot studies, particularly with spray application, could help decisions on treatment options.
- **Accumulation of phosphite in kauri** over time. A lack of knowledge regarding the ability of kauri to accumulate phosphite over time which will result in a higher concentration *in planta* than originally planned. This is unlikely to be a problem in kauri, but could be carefully monitored by residue testing.
- **Lack of knowledge of disease vectors.** What vectors other than pigs, humans and water runoff are important? Could these compromise the plots?
- **Poor vector control**. Pigs running through or wallowing in trial blocks could completely compromise any trials, as could unwanted human traffic. Pig control in the surrounding bush, fencing and signage (for humans and literate pigs) should help mitigate this. It is also important to limit researcher entry to essential tasks only, and restrict access to dry weather only.

- **Catastrophic events** For example, fire or flood. These can't really be controlled, but can be mitigated by having multiple trial sites.
- **Registration status of phosphite use in forests**. What is the status here? This is a grey area that should be clarified. Is ACVM registration required?

2.15 Feasibility

The trials necessary to prove the phosphite barrier principle in kauri forest are feasible. Although there will be a number of hurdles (certainly technical and possibly financial and political) that must be overcome, there is no obvious reason why the required trials are not technically feasible. There must be an acceptance at the start that the trials will be long term, perhaps over a period of 10 to 15 years, although some useful results are likely to be obtained earlier. With appropriate site selection, design, set-up, maintenance and assessment we can expect to get definitive results. We would anticipate being able to prove (or potentially disprove) the usefulness of phosphite barriers with some confidence. Without this information, authorities and forest guardians or managers would be unable to implement management systems with any confidence.

3 CONCLUSIONS

Trials investigating the efficacy of phosphite barriers for reducing the localised spread of *P. agathidicida* in forests will be long term, expensive and challenging to set up. Despite the problems, the work is still very important and worth doing. Work from overseas where phosphite barriers have been deployed to contain *Phytophthora* in native ecosystems, plus research in kauri forest showing the potential of phosphite in controlling *P. agathidicida* infection, provides evidence and confidence that the barrier technique will be beneficial.

Treating trees in advance of major symptom development, and taking it a step further and treating a band of potential hosts (phosphite barrier) in advance on an infection front, will be the most effective way of utilising phosphite as a tool for kauri dieback control. There is potential to protect important sites and to prevent or significantly slow the spread of the pathogen into new areas. The most difficult part will be providing the experimental proof of concept, which will be necessary to justify any future deployment of the technique.

A suggested implementation sequence is given below. This will need to be a multi-step process with re-evaluation points throughout. Once a project team is selected, and broad trial specifications are determined (based on the discussion in this report), the first requirement will be to scope, roughly map, and carry out preliminary soil sampling and testing of suitable trial sites. This will, in turn, determine most of the other trial criteria such as plot size, number of plots, potential assessments that should be made, and ultimately the cost of setting up trials. This part of the programme will take considerable effort, but is unpredictable until the work is done. There will need to be a degree of flexibility in project planning and contracting that allows for changes to be made as required, so that correct decisions can be made without compromising the project. This flexibility should continue throughout the project, with key re-evaluation or stop/go points written in. An oversight group, comprising the P&I team or other advisors might be a good way of managing this.

Other projects could potentially tie in with the phosphite barrier project. If plots are well set up they could be a mine of information in future years. But extreme care must be taken to avoid compromising the barrier work by excessive studies or movement on site. The project team must keep reminding themselves of the key question— *does a phosphite barrier slow the natural rate of spread of* P. agathidicida *through a site?*

3.1 Sequence of tasks

- Finalise key trial criteria, goals, assessments required.
- Assemble an appropriate project team.
- Identify potential sites, carry out site visits, including consultation and permission.
- Establish a broad trial plan and cost estimate (to be revised after site determination and mapping).
- Roughly maps sites and carry out the first round of soil tests to determine the disease front and trial site suitability.
- Produce a more detailed trial design and cost estimate.
- Carry out more detailed sampling to focus in on the disease front (following results from above).

- Produce a detailed design of the trial, mark out plots, mapping, transect establishment etc.
- Carry out a third round of sampling, focusing on key transects to accurately delineate infection boundaries at the trial starting point.
- Treatment application.
- Monitor phytotoxicity, perhaps one month and six months after treatment application.
- Monitor disease symptoms (6-months to yearly)
- Re-assess the infection front (focus on transects) every 1–2 years.
- Re-apply the barrier treatments.

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APPENDIX – SUMMARY OF LITERATURE AND OVERSEAS WORK

Very little has been published on using phosphite as a barrier to prevent *Phytophthora* spread. In terms of experimental proof of concept, the most thorough work is by Shearer, Crane and Fairman (2004), summarised below. A number of other authors have referred to the possibility of phosphite use as a barrier (Dunstan et al. 2009; Kanaskie et al. 2011; Pilbeam et al. 2000), but there has been little experimental evidence published. In discussing this point with other researchers there seems to be an understanding and acceptance that using phosphite as a barrier must be helping but that it's difficult, time consuming and expensive to prove experimentally. In spite of the minimal experimental proof, the phosphite barrier technique is being applied on a large scale in Western Australia to counter Phytophthora Dieback.

Kanaskie et al. (2011), in studies of sudden oak death (*Phytophthora ramorum*) in Oregon tanoak forests, commented … "One possible alternative to the current treatment approach is to create a host-free barrier well ahead of the leading edge of infestation. Given the scale of the Oregon infestation this would take years and would have considerable cost. Another option is to aerially apply a fungicide over large areas. The aim here is to prevent spread of *Phytophthora ramorum* either by protecting trees from infection or by reducing spore production in trees that become infected. …. Treatments that prevent infection of these tissues or reduce sporulation on them could be part of an integrated approach to controlling pathogen spread. An effective fungicide treatment could be used locally to prevent expansion of new infestations or to treat large areas of forest in advance of the disease, much like a host-free zone but at much lower cost."

The Australian Department of Environment and Conservation Phytophthora Dieback Guide (2012) discusses the use of phosphite in controlling disease spread. It states "…. phosphite can be used strategically for effective protection ahead of an advancing 'front' of *P. cinnamomi*. A 30–40-metre-wide swathe of phosphite can be applied in front of an advancing *P. cinnamomi* infestation to prevent root-to-root transfer of the pathogen across the barrier. If the infested area is upslope of the area to be protected the protective swathe would need to be wider than if it is downslope. This is because of the possibility of overland or subsurface transport of *P. cinnamomi* zoospores for considerable distances downslope following rainfall. In contrast, movement of an infestation upslope is generally slower, being mainly caused by root-to-root contact between plants."

Although guidelines for monitoring treatment effectiveness are presented in this ADEC Guide (e.g. marking of disease fronts, tagging of live and dead plants, plant censuses, photography, and use of control areas) there is no presentation of or referral to experimental evidence that such treatment is effective at slowing disease spread.

Dunstan et al. (2009) took an extreme approach in trying to eliminate relatively small infestations of *P. cinnamomi* in shrubland and heathland communities in Western Australia and Tasmania. Their approach included a combination of vegetation destruction, fungicides, fumigation and physical root barriers. The approach was successful in eradicating *Phytophthora* from these small infection foci, proving the concept and providing hope that recent small infection foci could be controlled in certain conditions, albeit at a high price. However, it is difficult to imagine this approach being either possible or successful in a kauri forest situation. It is interesting that Dunstan et al. didn't use phosphite in their eradication regime, stating that phosphite "inhibits but does not kill" and that it may "mask the presence of the pathogen while still producing infective stages". They added that "the use of phosphite should not be relied on

to contain the spread of *P. cinnamomi* and it should only be used as a temporary management tool, particularly where vegetation is already infested, until more long lasting and robust methods of control can be implemented."

Shearer, Crane and Fairman (2004) systematically investigated disease front extension and rate of extension of *Phytophthora cinnamomi* in a *Banksia* woodland in Western Australia. Specifically, they looked at the effect of trunk injection of trees combined with spraying of the understorey with various rates of phosphite, then followed the progression of the disease front under treated and untreated regimes. Compared to untreated controls, they found a significant reduction in the rate of disease spread, regardless of the phosphite regime.

Site: Banksia woodland, Swan Coastal Plain, Western Australia. Flat site.

Soil: coarse slightly acidic sand low in N, P and K.

Treatments: one untreated control and four different phosphite regimes, each including at least one foliar spray of understorey and trunk injection of trees. There were no treatments of either spray or trunk injection alone. Four replicates along a disease front.

Plots: 10 x 15 m plots, with one edge on the disease front and the remainder of the plot extending into the apparently healthy area

Site assessments: Soil minerals, organic carbon etc., structure.

Disease front assessments: *P. cinnamomi* infection expressed as death of a number of key indicator species. Paint used to mark the line of the disease front at each assessment. Measured 6 months, 4 years and 5 years after treatment.

Notes: Clearly visible disease front observed — sharp boundary between dead/dying plants and healthy. Fire through site one year after treatment.

Results: Phosphite treatment significantly reduced disease front extension at all three assessments. Minimal differences between the various phosphite regimes. Disease extension was reduced for up to 5 years. Disease extension rates were 1.3–1.8 m per year in untreated plots and the assumption is that this was predominantly through invasion/expansion on lateral roots of susceptible hosts.

Conclusion: "The large and significant reduction in disease extension between treated and non-treated plots suggests that phosphite application to disease fronts could control *P. cinnamomi* spread through roots and by root-to-root contact. High and low-volume spraying of foliage are the main methods of phosphite application currently being used in South-Western Australia to protect native threatened flora from *P. cinnamomi* infection. This study indicates that injection of the overstorey should accompany spraying of foliage to ensure long-lasting protection by phosphite."

Discussions with the Department of Parks and Wildlife, Western Australia

In correspondence with Sarah Barrett from the Department of Parks and Wildlife (DPAW) Western Australia, she indicated that they have very limited unpublished data on use of phosphite as a band along disease fronts. Many of the sprayed sites are monitored, but are typically a mosaic of infected and healthy sites. In one site near Albany WA, with aerially sprayed and control plots, disease spread has been monitored since 1996. They report good results with a significant reduction in spread. Sarah commented that monitoring of fronts is not simple, and getting good matching controls for comparison with treated areas can be difficult.

Typically fronts are convoluted, and good replication of trials is difficult, resulting in lots of small amounts of data, but nothing has been published. Sarah notes that they have monitored a number of sites where they have successfully reduced the spread of small infestations, even on steep slopes. In a site in the Fitzgerald River National Park, high intensity phosphite bark application used in a barrier method appears to have been effective — but no data has been published.

DPAW also implements treatment programmes utilising phosphite barrier techniques to contain Phytophthora dieback and to protect areas of value. Various criteria are scored to determine which sites are prioritised for treatment. Criteria include values such as importance of vegetation types, interference risks from vectors, size of area to be protected, and terrain (uphill or downhill from infected sites).

Commercial Phosphite Barrier Operations in Western Australia

In Western Australia, a company called Dieback Treatment Services (Managing Director Glenn Tuffnell) provides phosphite treatment services across the State (www.diebacktreatments.com). They work for both public and private parties, treating dieback-infected areas. Some of the work is in treating small infections on private properties, but much of their work is in treating large parkland areas with significant *P. cinnamomi* infections. In general, they are using the concept of the phosphite barrier to treat the margins of infected patches and carry out protective treatment to a band of vegetation in advance of the disease front. The treatment involves periodic (roughly every 3 years) injection of trees and spraying of understorey in a 15–20 m band along the disease front. At each treatment time, the disease margin is re-marked (by looking at indicator species), so the 15–20 m treatment band ahead of the disease front is maintained.

Trunk injection typically uses standard Chemjet tree injectors (the same as used effectively in kauri trials in NZ) to inject a 5% phosphite solution.

For understorey spraying, a 0.5% phosphite solution, amended with a surfactant is applied, using a motorised spray unit mounted on the back of a 4-wheel-drive vehicle. This vehicle carries a 1000 L tank, providing sufficient spray to treat a 20 m buffer 300 m long. A 200 m hose (up to 400 m?) facilitates access.

The estimated cost of the spray system (excluding the vehicle) is about AU$20,000. The full cost of the treatment operation (spray and injection) is about AU30-40 cents per m^2, depending on the forest type and density (Glenn Tuffnell personal communication).

It must be noted that most of the sites being treated in WA have relatively easy access by 4-wheel drive vehicle, at least to within a few hundred metres. The high pressure system with very long hose facilitates access. Access to many kauri dieback sites will not necessarily be so straight-forward.

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