

Date: June 2017
Reference: ND-TN002
Milestone: 2017-18 3.1.1

Technical Note

Results of detached needle assays from a field trial testing phosphite and copper for control of RNC on *Pinus radiata*

Authors: Carol Rolando, Stefan Gous, Nari Williams and Graham Coker

Corresponding author: Carol Rolando

Summary: Cuprous oxide may provide a reliable, short-term management option for red needle cast. A large field trial was carried out at Kaingaroa to test the efficacy of aerially applied foliar treatments for control of red needle cast (RNC) in *Pinus radiata*. Three rates of phosphite (6, 12 and 24 kg ha⁻¹, applied as FoschekTM) were tested, as well as cuprous oxide (applied as AgCopp 75) applied at 1.72 kg ha⁻¹ and diammonium phosphate (a fertiliser) applied at 6.6 kg ha⁻¹. All treatments were applied in 100 L ha⁻¹ water, except the control, which received no treatment. Three detached needle assays were conducted at approximately one month, three months and seven months after treatment application. One month after treatment application needles treated with cuprous oxide showed a significant reduction in the number of lesions formed (by 76%) and their length (by 87%) in comparison to the control. None of the other treatments significantly reduced lesion number or length in comparison to the control due to the high variability in infection responses. However, there was a trend of decreasing lesion length with increasing phosphite concentration up to a rate 12 kg ha⁻¹ phosphite. The application of diammonium phosphate at the equivalent of 6.6 kg ha⁻¹ did not significantly affect lesion length or number in comparison to the control. No significant differences between treatments were detected at the three and seven month assays.

Introduction

Red needle cast (RNC) is a foliar disease of *Pinus radiata* caused by *Phytophthora pluvialis* that has the potential to cause up to 16% annual growth loss in severely infected mature plantations. A cost-effective, FSC-compliant, chemical control strategy is needed to provide a short-term management option for control of severe outbreaks.

Several controlled trials on field and potted *P. radiata* trees known to be susceptible to RNC have been conducted to investigate the potential of fungicides for management of the disease (Rolando et al., 2014 a and b; Gous et al., 2015; Gous et al., 2016; Rolando et al., 2017 a and b). The results of these trials have shown that phosphite and copper provide potential as prophylactic treatments, despite a sometimes variable, and disappointing, response from phosphite treated and infected hosts (Rolando et al., 2017 b).

To increase our understanding of the performance of copper and phosphite applied as foliar treatments for management of RNC, it was necessary to test our initial findings at a field scale on mature *P. radiata* trees. Since little work had been conducted on the effects of foliar fertilization on disease development in

P. radiata this also presented an opportunity to test this in collaboration with the Growing Confidence in Forestry's Future programme.

The objectives of the trial described here were to:

1. determine the efficacy and persistence of aerially applied phosphite at three rates to control RNC in *P. radiata* using detached needle assays;
2. determine the efficacy and persistence of cuprous oxide and diammonium phosphate (DAP) as alternative treatments for RNC;
3. determine if there were any foliar phytotoxic effects of phosphite or phosphate to *P. radiata* when applied above label recommended rates;
4. investigate the links between tree and soil nutrition, tree growth, use of phosphite and phosphate and infection of *P. radiata* with *P. pluvialis* (link with GCFF programme);

Here we report on the results of the detached needle assays only (Objectives 1-3) and the preliminary implications for management. More detailed reports including outcomes on objectives 4 will follow as the results from the nutrition assays are made available.

Method

The trial was established at Kaingaroa Forest, Compartment 82/5 (1910886 E, 5732735 N NZTM), on a stand of *Pinus radiata* (seedlot 08/201) planted in 2009 (Figure 1). The site was ideal for an aerial spray trial as it was a large, contiguous flat area (Figure 1). Trees were on average 14.0 ± 0.35 m at the time of spraying.



Figure 1. Google Earth map of trial site

Three replications of six treatments (Table 1) were applied to the site in a randomised complete block design (RCBD) on 24 February 2016. All treatments were applied in 100 L ha^{-1} water using a Robinson 44 helicopter fitted with D6-45 nozzles (VMD $750 \mu\text{m}$). Four sub-plots of approximately 33 trees each were established within each plot.

exposed to the same solution *P. pluvialis* in a single flat-bottomed test-tube.

The variables lesion length and lesion number were analysed in SAS using mixed model procedures. Treatment (Treat) was included as a fixed effect with the random term reflecting the nested blocked design (Plot, Sub-plot and Tree). All data were appropriately transformed prior to analysis. For ease of interpretation all figures show untransformed data while tables of analyses show outcomes based on transformed data.

Results of detached needle assays

There was a significant effect of treatment on lesion length and number at the assay carried out one month after treatment application in early April 2016 (Table 2; Figures 2a and 3a). The most effective treatment was the application of cuprous oxide at 1.72 kg ha^{-1} , which resulted in both significantly fewer (by 76%) and smaller (by 87%) lesions than in the control. Due to the high variability in the response, none of the other treatments significantly reduced lesion length, although lesion length tended to decrease with phosphite treatment up to 12 kg ha^{-1} . The application of DAP at the equivalent of 6.6 kg ha^{-1} did not significantly affect lesion length or number in comparison to the control. Notably, the highest number of lesions were formed in this treatment one month after application (Figure 3a).

Table 1. Treatments applied to seven year old *P. radiata* trees. All treatments were applied in 100 L ha^{-1} water using a Robinson 44 helicopter fitted with D6-45 nozzles

No.	Treatment	Products
1	Untreated control	0
2	6 kg ha^{-1} phosphite ^a	15 L ha^{-1} Foschek
3	12 kg ha^{-1} phosphite ^a	30 L ha^{-1} Foschek
4	24 kg ha^{-1} phosphite ^a	60 L ha^{-1} Foschek
5	1.72 kg ha^{-1} cuprous oxide ^b + 80 ml Du Wett	2.3 kg ha^{-1} Cu_2O
6	DAP. @ 6.6 kg/ha of P	33 kg ha^{-1}

^a = Foschek™ (400 g/L phosphite, Zelam Ltd., Auckland);

^b = AgCopp 75, 75% cuprous oxide; American Chemet Corporation)

Detached needle assays

Detached needle assays were carried out at one, three and seven months after treatment application (April, June and October 2016). Three shoots were collected from the first five trees of each sub-plot for each assay. Six fascicles were selected from each tree and exposed to *P. pluvialis* as per standard practice for detached needle assays (Williams, 2013). The number of lesions per fascicle and their total length per needle was determined for each assay.

Analysis

The experiment was set up as a randomised complete block of three replications (Rep) of six treatments (Treat). Within each treatment plot (Plot) there were four sub-plots (Sub-plot) of five sampled trees (Tree). The variables total lesion length per fascicle (mm) and number of lesions were analysed at the tree level (experimental unit) as all fascicles from one tree were

Table 2. Results of the ANOVA for lesion length and number at 1, 3 and 7 months after treatment application.

Assay	Effect	DF (num/den)	F-value
Lesion length (mm)			
1 month	Treat	5/10	4.70**
3 month	Treat	5/10	2.36
7 month	Treat	5/10	1.51
Lesion number			
1 month	Treat	5/10	8.29**
3 month	Treat	5/10	3.31#
7 month	Treat	5/10	1.73

Note ** denotes significance at $P < 0.05$; # denotes $P = 0.05$

No significant differences were detected between treatments at the assays conducted three months (June 2016) and seven months (October 2016) after treatment application (Table 2), although the number and size of lesions in the cuprous oxide treatment was still notably smaller than that of the control, by 75% and 60% respectively. As with previous detached needle assays (Rolando et al., 2017b), this was partly a function of the high within-treatment variability obtained within each assay. At the seven month assessment lesion number and length was similar across the treatment set.

There were no signs of phytotoxicity in the trial, as would be indicated by severe yellowing or browning of crown foliage. Tree growth measurements were taken before treatment application and one year later. These assessments will be analysed together with the nutrition data and will provide a more definitive indication of the impacts of the phosphite and phosphate treatments on tree health, as indicated by growth.

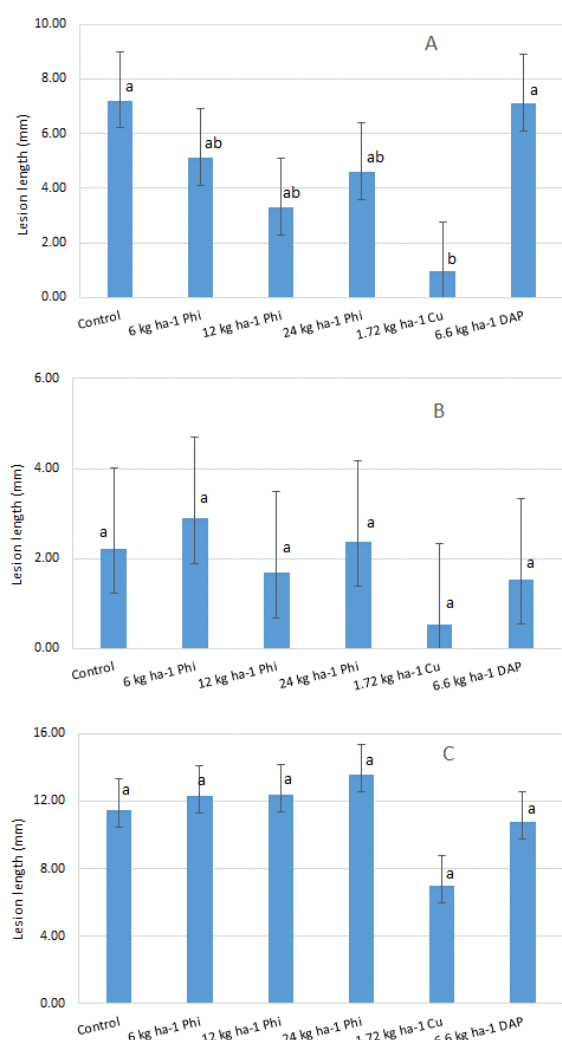


Figure 2. Average lesion length (mm) per detached fascicle collected from 7 year old *Pinus radiata* trees and inoculated with *Phytophthora pluvialis* at A) 1 month B) 3 months and C) 7 months after treatment application (see Table 1). Different lower case letters indicate statistically significant differences between chemicals. Bars indicate the standard error.

Implications of Results

The copper treatment was the most effective treatment reducing both lesion number and size at the one month assay as has been seen in previous trials (Rolando et al., 2017b, Gous et al., 2016; Gous et al., 2015). Unfortunately, good infection across all treatments was not obtained at the three month detached needle assay, making it difficult to interpret the outcome of this assay. However, given what is already known about this active ingredient, it would be unlikely that efficacy of copper would persist longer than three months after application (Gous et al., 2015; Rolando et al., 2017b).

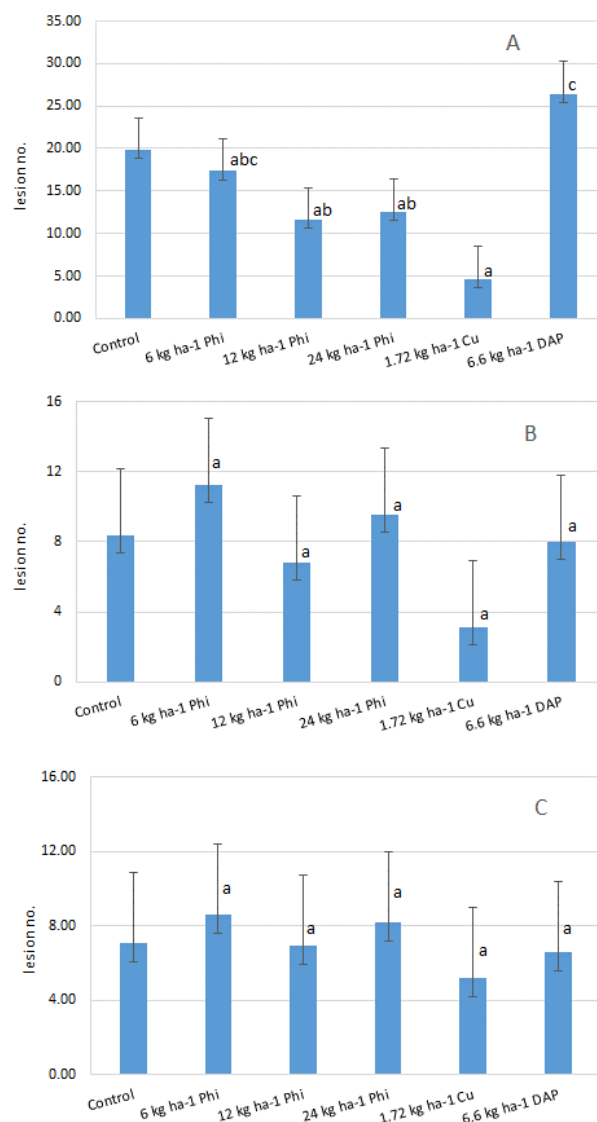


Figure 3. Average lesion number per tree from fascicles collected from 7 year old *Pinus radiata* trees and inoculated with *Phytophthora pluvialis* at a) 1 month b) 3 months and c) 7 months after treatment application (see Table 1). Different lower case letters indicate statistically significant differences between chemicals. Bars indicate the standard error.

None of the phosphite treatments appeared to provide a level of control that resulted in either significantly smaller or fewer lesions than where no treatment was applied, even at one month after application. Initial trials had indicated that suppression of lesion development in phosphite-treated trees had the potential to persist for up to one year after application (Rolando et al., 2013). However, contrary to initial indications, more recent trials focusing only on foliar application of this active ingredient indicate that phosphite may not be as effective a prophylactic treatment for management of RNC as previous trials have indicated. This outcome needs some consideration before further work with this active ingredient is considered.

The DAP treatment had no significant effect on lesion number or length. This trial indicates that such a fertilisation treatment would not be an effective option for management of red needle cast on similar site types.

A treatment with copper appears to be an effective option for management of RNC. The results from this trial indicate the efficacy of copper is unlikely to extend beyond three months, meaning that the timing and frequency of application is going to be an important determinant of effective control. The sensitivity of *P. pluvialis* to copper ions has yet to be determined, but is the subject of a current dose response trial.

The results presented here capture only the outcome of the detached needle assays for this trial. A more comprehensive report exploring the host x treatment x site nutrition interaction in depth is in preparation.

Acknowledgements

Funding for this research came from the Needle Disease Strategy, the Growing Confidence in Forestry's Future and the Healthy Trees Healthy Future research programmes. Both of the latter programmes are jointly funded by the Ministry of Business Information and Employment (MBIE) and the Forest Growers Levy Trust, with the support of the NZ Forest Owners Association (FOA) and the NZ Farm Forestry Association (FFA). The authors would like to thank Catherine Banham, Caro Gous, Liam Wright and Rodrigo Osorio for the technical support on all the logistics of collecting foliage samples in-field and conducting the detached needle assays in the laboratory.

References

Gous, S.F., Rolando, C.A., Bader, M K-F, Williams, N. (2015). Evaluation of foliar copper to control red needle cast disease in *Pinus radiata*. Client Report for Needle Disease Strategy. Scion, Rotorua, New Zealand.

Gous, S.F., Rolando, C.A., Bader M K-F, and Todoroki, C. (2016). Evaluation of the persistence of cuprous oxide (operational Dothistroma treatment) to control and manage RNC 2015/2016. Client Report for Needle Disease Strategy. Scion, Rotorua, New Zealand.

Rolando, C.A., Williams, N.W. and Bader, M. (2013). Efficacy and persistence of phosphite for control of RNC in *Pinus radiata*. Client report for Needle Disease Strategy. Scion, Rotorua, New Zealand.

Rolando, C.A. Williams, N.W. and Bader, M. (2014a). Efficacy and persistence of phosphite for control of RNC in *Pinus radiata*. Client Report for Needle Disease Strategy. Scion, Rotorua, New Zealand

Rolando, C. A., Gaskin, R., Horgan, D. Williams, N. and Bader, M. (2014b). The use of adjuvants to improve uptake of phosphorous acid applied to *Pinus radiata* needles for control of foliar *Phytophthora* diseases. New Zealand Journal of Forestry Science: 44(8).

Rolando, C. A., Williams, N.W., Gardner, J.A. Dick, M.A. and Bader, M K-F (2017a). Chemical control of two *Phytophthora* species infecting the canopy of Monterey Pine (*Pinus radiata*). Forest Pathology: 1-10.

Rolando, C.A. Gous, S.F., Bader, M K-F, and Williams, N.W. (2017b). Pot trial to evaluate the efficacy and persistence of phosphite and cuprous oxide to control RNC. Client Report for Needle Disease Strategy. Scion, Rotorua, New Zealand

Williams, N. (2013). Red needle cast SOP. Scion Internal Report No. 51194. Scion, Rotorua, New Zealand.