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Quantification of root phosphite concentrations for evaluating the potential of foliar phosphonate sprays for the management of avocado root rot

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ABSTRACT

In South Africa, phosphonate trunk injections are widely used in a preventative management strategy against avocado root rot caused by *Phytophthora cinnamomi*. Due to increasing costs, alternative application methods must be investigated. The efficacy of different phosphonate foliar spray treatments was evaluated in two trials that were each situated in a climatically different region. Efficacy was evaluated through quantification of root phosphite (breakdown product of phosphonates) concentrations at different time points, following fall and summer applications. Since no high-throughput cost-effective analytical methods are available for phosphite quantification from avocado roots, a phosphite extraction and purification method was first developed, from which phosphite was quantified using a publically available liquid chromatography-mass spectrometry (LC-MS/MS) method. Foliar potassium phosphonate sprays, applied as three weekly sprays (full- and ¾ volume sprays) in fall, did not result in significantly lower root phosphite concentrations (8, 12 and 23 weeks after application) than the trunk injection. This was also true for two potassium phosphonate foliar sprays applied in summer (8 and 14 weeks after application) in the one trial. However, in the other trial, the summer applied potassium phosphonate foliar sprays had significantly lower root phosphite concentrations than the trunk injection. Ammonium phosphonate foliar sprays, three sprays applied in fall and two in summer, consistently yielded higher or similar root phosphite concentrations than the trunk injection. The ammonium phosphonate foliar sprays furthermore yielded significantly higher root phosphite concentrations than the corresponding potassium phosphonate foliar spray treatment. This was true for almost all time points, except 8-weeks after the summer application in one trial. Phosphite fruit residues were significantly higher for the foliar spray treatments than for the trunk injection in the one trial, but in the other trial it was similar or lower.

1. Introduction

Avocado root rot caused by *Phytophthora cinnamomi* is effectively managed using phosphonate fungicides (salts and esters of phosphite [syn. phosphonic acid]) world-wide, including South Africa (Darvas et al., 1984; Pegg et al., 1987). In South Africa, the pathogen previously caused wide-spread destruction in orchards. This changed when Darvas et al. (1984) discovered fosetyl-aluminium (alkyl phosphonate) trunk injections, which was subsequently also registered in South Africa. In addition to fosetyl-aluminium trunk injections, potassium phosphonate has also been registered in South Africa as a trunk injection for preventative- and curative root rot management. This product is currently widely used due to its cost-effectiveness compared to fosetyl-aluminium. In addition to potassium phosphonates, ammonium phosphonate is also available in South Africa as a registered fungicide on crops other than avocado.

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Initially, phosphonate management of avocado root rot was focused on the curative treatment of declining trees. However, as the health of declining trees improved, the focus has since moved to preventative phosphonate management strategies (Whiley et al., 1995). In South Africa, a preventative management strategy consists of two potassium phosphonate trunk injections applied annually, one in fall (after the summer flush hardened off) and another in summer (after the spring flush hardened off). These application windows are very effective due to root flushing occurring during these time points, and the source sink translocation of phosphonates (Whiley et al., 1995). The highly mobile nature of phosphonates in plants unfortunately also results in translocation to fruits, and exceedances of fruit residue limits. This was not problematic during the first registrations of phosphonates in the 1980's, since there were no set maximum residue levels. Consequently, residue data were not a requirement for product registration. However, this situation changed in 2014, when the European Union started to enforce residue limits (50 mg/kg) for phosphonate products on avocado and several other crops. The European Union is the largest avocado export market for South Africa.

Due to increasing labor costs, and trunk injections possibly causing damage to tree trunks, alternative application methods are required for replacing phosphonate trunk injections that are widely used in South Africa. In South Africa, labour cost in the agriculture sector increased drastically in 2013 by approximately 50% due to labour unrest (Pahle, 2015). This has resulted in alternative application methods such as foliar sprays becoming more cost effective. Currently, approximately four 0.5% (a.i. phosphorous acid) foliar sprays are almost comparable in cost to two trunk injections in new high density orchards (unpublished data). In contrast, in 2001 only two foliar sprays were similar in cost to two trunk injections (Duvenhage, 2001). In addition to increasing costs, another negative aspect of trunk injections is that it can cause damage to the trunk wood with prolonged use (Robbertse and Duvenhage, 1999). Trunk sprays containing penetrants are an alternative application method that is effective on young avocado trees with green stems (Giblin et al., 2007), and on some threatened native plant species in some countries (Crane and Shearer, 2014; Dunstan and Hardy, 2005; Garbelotto et al., 2007). However, this application method is not effective for older bearing avocado trees (Giblin et al., 2007). A better alternative application method is foliar phosphonate sprays that were first registered in Australia as 0.1% (a.i. phosphorous acid) foliar potassium phosphonate sprays. However, these were later found ineffective by growers. Therefore, emergency use permits were obtained for 0.5% (a.i. phosphorous acid) foliar sprays, which are still being used in Australia (Whiley et al., 2001; personal communication, Elizabeth Dann, University of Queensland, Brisbane, Australia). The number of sprays required is not well defined, with the emergency use registration stating a limit of no more than five sprays. Whiley et al. (2001), in a non-peer reviewed article recommended applications of between three to eight 0.5% foliar sprays. The variable number of recommended foliar sprays might be due to differences in season, location and crop load (Whiley et al., 2001). Therefore, in Australia, it is recommended that growers monitor their root phosphite (breakdown product of phosphonates in plants) levels through a commercial laboratory to determine the number of sprays required (Whiley et al., 2001).

In South Africa, limited work has been conducted on the efficacy of foliar phosphonate sprays. The studies have all only been published in non-peer reviewed journals. Duvenhage (2001) evaluated the efficacy of two 0.75% (a.i. phosphorous acid) potassium phosphonate foliar sprays (one after summer flush completion and the other after spring flush completion) in one orchard, and reported that these were effective. McLeod et al. (2015) evaluated the efficacy of three to four foliar potassium phosphonate sprays applied at different concentrations (0.5%, 0.75% and 1% a.i. phosphorous acid) in two orchard trials. None of the foliar sprays were effective in comparison to the registered trunk injection. This was most likely due to the fact that foliar sprays were applied with a knapsack sprayer, which resulted in too low spray volumes being applied.

The efficacy of phosphonate foliar sprays is known to be influenced by spray volume, but limited information is available. In native threatened plant communities in Australia, high volume aerial foliar sprays were shown to be more effective than low volume sprays (Shearer et al., 2012). A similar finding has been reported in avocado in Australia where high spray volumes were more effective. This is most likely due to the fact that more active ingredient is applied to trees (Whiley et al., 2001). The spray volume specified by the emergency use label for potassium phosphonates in Australia states "apply spray volume of 2000–3000 L/ha for matures trees (depending on tree size)". This is a rather wide range to select from, which may lead to suboptimal or inconsistent results. In deciduous fruit crops in South Africa, spray volume is determined using the Unrath tree-row-volume (TRV) model. This model could be useful for determining spray volumes for the application of foliar phosphonate sprays to avocado trees. The Unrath model calculates a high spray volume using the formula: $\textit{Spray volume} = \frac{\text{tree height} \times \text{tree canopy diameter} \times 1200}{\text{row width}}$. The constant in formula (1200) can vary according to tree crop type (Unrath et al., 1986).

In plants, phosphonate dissociates into anions, hereafter referred to as phosphite, which is important in plant tissue for pathogen suppression. This, however, may vary in different *Phytophthora* host plant systems. In general, a negative linear relationship has been reported for lesion length development and phosphite plant tissue concentration for threatened native Australian plant species that are effectively controlled with phosphonates. However, this is not true for species where phosphonates are less effective (Shearer and Crane, 2009; Shearer et al., 2012; Wilkinson et al., 2001a). El-Hamalawi et al. (1995) also found a negative linear relationship between bark phosphite concentration and inhibition of *P. citricola* in avocado. Smillie et al. (1989) furthermore reported a close correlation between the concentration of phosphite present at the site of inoculation and the extent of protection against *P. cinnamomi, P. nicotianae* and *P. palmivora* inoculated onto phosphonate treated lupine, tobacco and pawpaw plants respectively.

Knowledge on phosphite concentrations in plants can be useful for optimizing phosphonate application methods and dosages. In peer reviewed literature, this has been done for the management of *P. cinnamomi* in threatened native plant species in Australia (Crane and Shearer, 2014; Fairbanks et al., 2000; Shearer and Crane, 2009; Shearer et al., 2012), *P. citrophthora* in citrus (Schutte et al., 1991), *P. citricola* canker development in avocado (El-Hamalawi et al., 1995) and *P. cinnamomi* avocado root rot (Ouimette and Coffey, 1989). A few non-peer reviewed articles have evaluated foliar phosphonate sprays based on root phosphite concentrations in avocado in Australia (Whiley et al., 2001) and South Africa (Duvenhage, 2001; McLeod et al., 2015). Quantification of root phosphite is especially useful for optimizing phosphonate applications for a preventative management strategy, since the pathogen is absent, or present at very low levels and does not cause enough damage, or do so inconsistently within orchards. Foliar sprays, in general, cannot be evaluated effectively on diseased trees since declining trees do not have sufficient foliage for the uptake of foliar applied phosphonates (Darvas, 1983).

A number of analytical methods have been published for quantifying phosphite in different plant tissues including radiolabelling, gas chromatography, gas chromatography – mass