EVALUATING AMMONIUM- AND POTASSIUM PHOSPHONATE FOLIAR SPRAYS OVER TWO SEASONS BASED ON ROOT PHOSPHITE CONCENTRATIONS FOR THE PREVENTATIVE MANAGEMENT OF PHYTOPHTHORA ROOT ROT

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ABSTRACT

Phosphonates are widely used for managing Phytophthora root rot on avocado. In South Africa, only trunk injections are employed for the preventative and curative management of the disease, whereas in other regions of the world foliar sprays are also used for preventative disease management. Trunk injections are becoming increasingly expensive due to a constant increase in labour cost. Therefore, the first aim of the current study was to determine whether ammonium phosphonate (Brilliant®) and potassium phosphonate (Fighter®) foliar sprays evaluated in five orchard trials over two seasons yielded root phosphite (breakdown product of phosphonates) concentrations that were comparable to the registered potassium phosphonate trunk injection (Avoguard[®]) and registered alkyl phosphonate foliar sprays (Aliette[®]). In the 2018/19 season, in the Mooketsi and Letaba production regions, four or five Brilliant® or Fighter® foliar sprays yielded root phosphite concentrations that were comparable to those obtained with the trunk injection treatment (preventative dosage). In the same season, a trial in Howick that evaluated five Brilliant® or Fighter® foliar sprays yielded similar results. In the 2019/20 season, four Brilliant® or Fighter® sprays evaluated in Mooketsi and Letaba also yielded root phosphite concentrations that were comparable to the trunk injection treatment (curative dosage). In both seasons, the Aliette® foliar sprays yielded the lowest root phosphite concentrations, which were sometimes not different from the untreated control. A second aim of the study was to determine if Fighter® and Brilliant® foliar sprays would result in exceedances of the European Union maximum residue level (MRL) for fosetyl-Al (50 mg/kg). None of the Brilliant[®] and Fighter[®] spray treatments resulted in exceedances of the MRL. The data obtained in this study will contribute toward the registration of Fighter® and Brilliant® as foliar sprays on avocado for the preventative management of Phytophthora root rot in South Africa.

INTRODUCTION

Phosphonate fungicides are widely used for managing Phytophthora root rot in avocado in a preventative or curative manner (Darvas *et al.*, 1984; Pegg *et al.*, 1987). Various phosphonate formulations are available, including potassium-, sodium-, ammoniumand alkyl phosphonates. All these formulations ultimately break down to phosphonic acid and phosphite (also referred to as phosphonate anion, hydrogen phosphonate). Phosphite anions are the active compound involved in suppressing oomycete pathogens (Guest and Grant, 1991; Dann and McLeod, 2021) including the Phytophthora root rot-causing pathogen, *Phytophthora cinnamomi*.

Due to the high mobility of phosphite in plants, with translocation occurring in the xylem and phloem, various phosphonate application methods are effective. In avocado, trunk injections and foliar

sprays are the two most widely used application methods. In South Africa, potassium phosphonate (Fighter® and Avoguard®) trunk injections are registered and widely used for preventative and curative management of Phytophthora root rot. Although alkyl phosphonate foliar sprays (Aliette®) are registered in South Africa, these are not used commercially on bearing trees due to high product cost. In Australia and the USA, potassium phosphonate foliar sprays and trunk injections are also registered application methods. In Australia, foliar sprays are widely used as a preventative treatment against root rot. Only 0.5% a.i. (phosphorous acid) foliar sprays are recommended since 0.1% a.i. sprays were found to be ineffective (Whiley et al., 2001). According to the label of Agriphos 600[®], no more than five annual sprays may be applied (https://portal.apvma.gov.au).

Root phosphite concentrations are used in Australia for evaluating the efficacy of phosphonate treatments and to determine the number of foliar sprays required (Thomas, 2001; Thomas, 2008). Phosphonates are mainly applied in autumn (after the summer flush has hardened off), since this is the longest root flush window, with additional sprays being applied in summer (after the spring flush has hardened off) if root phosphite concentrations start being depleted. The number of foliar sprays required can vary among production regions, for example in some regions with a warm climate and vigorously growing trees more sprays are required than in regions with less vigorously growing tees (https://youtu. be/0T2Kz5tNfX0). Initially, in Australia, the maintenance of a critical root phosphite concentration of at least 25 μ g/g was recommended for effective disease control (Thomas, 2008). This, however, has recently been increased to 80 μ g/g (https://youtu. be/0T2Kz5tNfX0). Since root phosphite concentration decreases over time, a substantially higher root phosphite concentration is required just after phosphonate application to prevent root phosphite concentrations from dropping below the critical level prior to re-application.

The aim of the current study was to conduct orchard trials over two seasons (2018/19 and 2019/20) to determine whether root phosphite concentrations achieved with potassium- (Fighter®) and ammonium phosphonate (Brilliant®) foliar sprays are comparable to those obtained with a registered potassium phosphonate trunk injection (Avoguard®) and alkyl-phosphonate foliar sprays (Aliette®) in South Africa. Since South Africa's largest export market, the European Union, enforces a maximum residue level (MRL) of 50 mg/kg for fosetyl-Al, fruit residues were also determined in the trials.

MATERIALS AND METHODS Orchard trials

In the 2018/19 season, three trials were conducted that were situated in the Howick, Mooketsi and Letaba production regions. In the 2019/20 season, two trials were conducted, one each in the Mooketsi and Letaba production regions. Details of the orchard sites are provided in Table 1, as well as the foliar spray volumes that were applied. The orchard trees did not exhibit any foliar symptoms of Phytophthora root rot, since phosphonates were evaluated for use in a preventative management strategy. Treatments and application dates applied in each of the five tri-

Trial site parameters and foliar spray volumes	Trial number and information			
2018/19 season	Mooketsi trial	Letaba trial	Howick trial	
Cultivar/rootstock	Maluma-Hass/Duke 7	Hass/Duke 7	Hass/Dusa	
Row width and intra-row tree spacing	7 m x 3.5 m	7 m x 3.5 m	7 m x 4 m	
Planting density	408	408	357	
Tree age	11 years	7 years	6 years	
Tree height and canopy diameter	3.9 m x 4.8 m	3 m x 4.1 m	5 m x 5 m	
Irrigation type	Sprinkler	Sprinkler	Microjets	
Soil type	Sandy loam	Clay loam	Hutton	
Foliar spray volume applied	2100 L/ha	1500 L/ha	2974 L/ha	
2019/20 season	Mooketsi trial	Letaba trial		
Cultivar/rootstock	Maluma-Hass/Duke 7	Hass/Duke-7		
Row width and intra-row tree spacing	7 m x 3.5 m	7 m x 3.5 m		
Planting density	408	408		
Tree age	12 years	8 years		
Tree height and canopy diameter	3.9 m x 4.8 m	3.0 m x 4.10 m		
Irrigation type	Sprinkler	Sprinkler		
Soil type	Sandy loam	Clay loam]	
Foliar spray volume applied	2100 L/ha	1500 L/ha]	

 Table 1: Information of orchards that were used to evaluate phosphonate foliar sprays (Fighter[®] and Brilliant[®]) in the 2018/19 and 2019/20 seasons

 $\label{eq:Table 2: Phosphonate treatments and application dates for avocado orchard trials conducted in the 2018/19 and 2019/20 seasons$

Treatment	Autumn application dates	Summer application dates	Autumn application dates	Summer application dates
2018/19 Season	Mooketsi trial		Letaba trial	
Untreated control	None	None	None	None
4 x 0.5% Fighter®	28 March 2018,	None	9, 16 &	None
foliar spray	4, 11 & 18 April 2018		25 April 2018,	
. ,			4 May 2018	
5 x 0.5% Fighter [®]	28 March 2018,	None	9, 16 &	None
foliar spray	4, 11, 18 &		25 April 2018,	
	23 April 2018		4 & 9 May 2018	
4 x 0.5% Brilliant®	28 March 2018,	None	9, 16 &	None
foliar spray	4, 11 & 18 April 2018		25 April 2018,	
5 x 0 5% Brilliant®	29 March 2019	Nono	4 May 2018	Nono
foliar spray	20 March 2010, 4 11 18 &	None	25 Anril 2018	None
	23 April 2018		4 & 9 May 2018	
Trunk injection 0.3a a i $/\text{m}^2$	4 April 2018	4 December 2019	16 April 2018	4 December 2018
Aliette® foliar spray	None	11 Sentember	None	11 Sentember
preventative (every	None	25 October	None	25 October
6-weeks)		3 December		3 December 2018
o weeksy		2018, 15 January.		15 January.
		26 February,		26 February,
		9 April 2019		9 April 2019
	Howick trial			
Untreated control	None	None		
5 x 0.5% Fighter [®]	28 July,	None		
foliar spray	5, 10 & 24 August,			
	3 September 2018			
5 x 1% Fighter®	28 July,	None		
foliar spray	5, 10 & 24 August,			
5 x 0 5% Brilliant®	3 September 2018	None		
foliar spray	5 10 & 24 August	None		
ional spray	3 September 2018			
5 x 1% Brilliant®	28 July,	None		
foliar spray	5, 10 & 24 August,			
. ,	3 September 2018			
Trunk injection 0.5 g a.i./m ²	31 July 2018	1 November 2018		
Aliette [®] foliar spray	None	15 December		
preventative (every		2018,		
6-weeks)		29 January &		
		12 March 2019		
2019/20 season	Mooketsi trial		Letaba trial	
Untreated control	None	None	None	None
foliar spray	4, 9 & 20 April, 2 May 2019	None	2 & 6 May 2019	None
4 x 0.5% Brilliant [®]	4. 9 & 26 April.	None	10 & 26 April	None
foliar spray	2 May 2019		2 & 6 May 2019	
Trunk injection 0.5 g a.i./m ²	2 May 2019	2 November 2019	2 May 2019	2 November 2019
Aliette [®] foliar sprav curative	None	12 September,	None	12 September,
(every 4-weeks)		10 October,		10 October,
		1 November,		1 November,
		5 December 2019		5 December 2019
		2 January,		2 January,
		30 January,		30 January,
		27 February,		27 February,
		26 March 2020		26 March 2020



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als are shown in Table 2. In each of the trials, each of the treatments were replicated six times, using a completely randomized block design. Each replicate consisted of eight trees, with a buffer row between treatment rows. Potassium phosphonate (Fighter[®]) and ammonium phosphonate (Brilliant[®]) foliar sprays were applied as 0.5% a.i. (phosphorous acid) sprays, for which the pH was adjusted to 7.2 using potassium hydroxide. For Fighter 350[®] a 0.5% a.i. spray consisted of 1.42 L product/100 L and for Brilliant[®] 1.67 L product/100 L. At the Howick trial, 1% a.i. Fighter[®] and Brilliant[®] sprays were also evaluated to determine phytotoxicity of the sprays in this region. Phytotoxicity trials in the Mooketsi and Letaba regions were conducted previously (Masikane et al., 2018). Spray volumes required per hectare were determined using the tree-row-volume (TRV) Unrath formula (Unrath, 1986):

Spray volume = tree height x tree canopy diameter x 900 row width

Foliar sprays in the Mooketsi and Letaba region were applied using axial fan sprayers, whereas in the Howick trial a Cima sprayer was used.

Avoguard[®] injections and Aliette[®] foliar sprays were applied according to the registered label recommendations. Depending on the trial, for the injections a preventative dosage (0.3 g a.i./m²) or curative dosage $(0.5 \text{ g a.i.}/\text{m}^2)$ was employed (Table 2). For the Aliette[®] foliar sprays, preventative sprays (6-weekly) or curative sprays (4-weekly) were applied depending on the trial.

Root sampling and root phosphite concentration analysis

Feeder root samples were collected from the four centre trees in each replicate at three (2018/19 season) or two (2019/20 season) time points post-application (Figs. 1 and 2). The post-application time point refers to the number of weeks after the autumn trunk injection was applied in the trials. Root phosphite was extracted and quantified as previously described (McLeod et al., 2018), using a few modifications.

Fruit residues

Fruit harvested from the four centre trees, approximately 2 kg, in each replicate was sent for fosetyl-Al analysis to an accredited laboratory (Hearshaw and Kinnes Analytical Laboratory (Pty) Ltd, Tokai, South Africa).

RESULTS AND DISCUSSION Root phosphite concentration analysis

In all of the trials, there was a marked decrease in root phosphite concentrations just after application (10- or 15-weeks post-application) and at the last measured time point (46- or 47-weeks post-application) (Figs. 1 and 2). This was expected, since root phosphite concentrations are known to decline following application. This can be due to a dilution effect occurring with tree growth, fruit removal, leaf



2018/2019 Season Mooketsi

10 w (June'18) = 29 w (Oct'18) = 47 w (March '19)

A

350

Figure 1: Effect of different phosphonate treatments on root phosphite concentrations measured at different time points post-application (15 to 47 weeks) in avocado orchard trees in the 2018/19 season. The trials were conducted in three production regions (Letaba, Howick and Mooketsi).

abscissions and a low level of leaching of phosphite from roots (Dann and McLeod, 2021). The root phosphite concentrations obtained 10- to 15-weeks post-application in the five trials were much lower or close to the critical root phosphite concentration $(80 \ \mu g/g)$ used in Australia. Across the five trials, the 10- to 15-week root phosphite concentrations varied from 51 to 101 μ g/g for the Fighter[®] sprays, 64 to 208 μ g/g for the Brilliant[®] sprays, 45 to 84 μ g/g for curative trunk injections and 33 to 36 μ g/g for preventative trunk injections (Figs. 1 and 2). Considering that these concentrations declined substantially from the 10- to 15-weeks to the 46or 47-weeks post-application time point, the root phosphite concentrations in our trials seem insufficient for pathogen control based on the information from Australia. The seemingly low root phosphite concentrations obtained in our study could be due



Figure 2: Effect of different phosphonate treatments on root phosphite concentrations measured at two time points post-application (15 to 47 weeks) in avocado orchard trees in the 2019/20 season. The trials were conducted in two production regions (Letaba and Mooketsi).

to differences in the root phosphite quantification method utilised in our study and in Australia. In Australia, root phosphite analysis is conducted by a commercial laboratory and the method that is used is thus not available publicly. It is furthermore possible that the critical root phosphite concentration required for suppression of *P. cinnamomi* may vary according to rootstock susceptibility to the pathogen. In South Africa, clonal moderately tolerant 'Duke-7' and highly tolerant clonal 'Dusa'® rootstocks are widely utilised, which may require lower root phosphite concentrations than susceptible 'Reed' seedling rootstocks that are used in Australia. In scientific peer-reviewed literature, no information is available for any plant species regarding a critical root phosphite concentration required for suppression of oomycetes.

In the 2018/19 season, root phosphite concentrations obtained with the four or five Fighter[®] and Brilliant[®] foliar sprays were higher than the preventative trunk injection treatment (0.3 g a.i./m²), except for the 4 x Fighter[®] spray treatment at the Howick trial that was similar to the trunk injection treatment (curative 0.5 g a.i./m²) (Fig. 1). The Fighter[®] and Brilliant[®] foliar sprays yielded root phosphite concentrations at the last sampling time point (46 weeks post-application) that were comparable to that obtained with the trunk injection. The Aliette[®] foliar sprays yielded the lowest root phosphite concentration of all the phosphonate treatments, which were sometimes not much different from the untreated control. Although Aliette[®] applied as a foliar spray is registered in South Africa, it is not used commercially and its efficacy under commercial conditions is thus unknown. The low root phosphite concentrations obtained

with the Aliette® foliar sprays are not unexpected considering the dosage applied and time of application. The Aliette® sprays were applied at a relatively low dosage (0.19% a.i.) during six (preventative dosage) to eight (curative dosage) sprays, thus resulting in a total of 1.14% and 1.52% a.i. being applied annually, respectively. In contrast, the Brilliant[®] and Fighter[®] sprays were applied at 0.5% a.i. as four or five sprays, thus resulting in a total annual application of 2% and 2.5% a.i. respectively. The time of application specified by the Aliette® label also likely contributed further towards low root phosphite concentrations, since the label specifies that applications must be made from after the spring flush has hardened off (usually September/October) at 4-week (preventative) or 6-week (curative) intervals with the final spray being applied in April. This resulted in several of the sprays being applied when trees were bearing small fruits. Due to the source-sink translocation of phosphite, phosphite will also be translocated to the small fruit (Whiley et al., 1995). More translocation of phosphite to fruit with the Aliette® sprays than for the Brilliant[®] and Fighter[®] sprays is evident from the fosetyl-Al fruit residues of the Aliette® treatment being comparable with the four Fighter[®] sprays (Fig. 3). This is irrespective of the fact that the amount of active ingredient applied annually for Aliette® was lower than that of Fighter[®].

The presence of phosphite in the roots of untreated trees was unexpected, since the trees were not treated for 1-year (2018/19 season) or 2-years (2019/20 season) before initiation of the trials. This suggests that phosphite is stored in trees as previously hypothesized by Dann and McLeod (2021).

In the 2019/20 season, four Brilliant[®] or Fighter[®] sprays were evaluated (Fig. 2). In both trials, these treatments yielded root phosphite concentrations that were comparable to the curative trunk injection treatment (0.5 g a.i./m²) at both of the measured time points (10and 46-weeks). The exception was for the Brilliant[®] treatment at the Mooketsi trial. Similar to the 2018/19 season, the Aliette[®] foliar sprays yielded the lowest root phosphite concentrations.

Fruit residues

In both seasons, considering all five trials, the Fighter[®] and Brilliant[®] foliar sprays did not result in exceedances of the MRL (Fig. 3). This is in agreement with a previous report for two trials conducted in the 2017/18 season (Masikane *et al.*, 2018). In the 2019/20 Letaba trial, the curative trunk injection treatment exceeded the 50 mg/kg MRL. Fruit from the untreated control trees also contained fosetyl-Al residues, as was observed for root phosphite.

CONCLUSION

Brilliant[®] and Fighter[®] foliar sprays (four or five sprays) were as effective as the registered trunk injection based on root phosphite concentrations. The registered Aliette® foliar sprays were ineffective based on root phosphite concentrations, most likely due to the low annual dosage applied and the time of application. When applying Brilliant® and Fighter® foliar sprays it is important to use the correct spray volume based on the Unrath formula, since too low spray volumes will result in inefficacy (unpublished data). It is furthermore important to apply the sprays as soon as possible after harvest, since P. cinnamomi activity in soil and roots most likely increases around the harvesting period (Jolliffe, 2019; unpublished data), and root phosphite concentrations are being depleted. This makes the use of foliar sprays in orchards with late hanging fruit challenging, since previous trials showed that applications of Brilliant® and Fighter® foliar sprays to late hanging fruit can result in increased fruit residues 21-days post application. This was more problematic in the Letaba trial than in the Mookesti trial (McLeod et al., 2020). Due to the high dosage of Brilliant[®] and Fighter[®] applied (0.5 g a.i.), it is important to adjust the pH of spray solutions to 7.2 using KOH after the products have been mixed in the spray tank to prevent foliar burn. It is furthermore advised to spray in the morning and not when extreme heat conditions prevail. Applications for the amendment of the Brilliant[®] and Fighter[®] labels to include foliar spray application in avocado must still be submitted to the Registrar.

Acknowledgements

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B 2018/2019 Season Howick 12 10 Fosetyl-Al (mg/kg) 8 6 4 2 0 Control 5x Fighter® 5x Fighter® 5x Brilliant® 5x Brilliant ® Injection Aliette® (1% a.i.) (0.5% a.i.) (1% a.i.) (0.5 g foliar (0.5% a.i.) a.i./m2)





Figure 3. Effect of different phosphonate treatments on fosetyl-Al fruit residues in the 2018/19 season (A and B) and 2019/20 season (C) in different avocado production regions (Mooketsi, Letaba and Howick).

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