

# The use of agricultural adjuvants to improve fungicide spray deposition on grapevine leaves

Sybrand A. van Zyl<sup>1,2</sup> and Paul H. Fourie<sup>1,3</sup>



<sup>1</sup> Department of Plant Pathology, Private Bag X1, University of Stellenbosch 7602

<sup>2</sup> Present address: ExperiCo (Fruit Technology Solutions), P O Box 4022, Idas Valley, Stellenbosch 7609

<sup>3</sup> Citrus Research International, P O Box 28, Nelspruit 1200

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## Introduction

The primary objective of spray technology is to optimise deposition and activity of fungicide or pesticide active ingredients, of which the plant surface is a critical component in the spray application process, specifically the retention of spray droplets (Wagner et al., 2003). Good deposition of active ingredient on the target site is an essential requirement for effective pest and disease management (Brink et al., 2004, 2006). Target sites for most fungicide or pesticide spray applications are fruit or leaves, where the cuticle covers the outer plant surfaces. Adjuvants are often used for spreading and retention of the active ingredients of agrochemicals onto the water repellent cuticular waxes (Bargel et al., 2006), especially when contact fungicides are applied (Holloway, 1970; Holloway, 1993; Wagner et al., 2003).

Waxy leaf surfaces are readily wetted by aqueous sprays containing a suitable surface-acting-agent (Stevens et al., 1993). Adjuvants are additives with specific surface-acting-agents (surfactants), commonly applied along with pesticides to improve spray performance (Ryckaert et al., 2007). These include features such as deposition and retention (De Ruiter et al., 1990; Hall et al., 1993, 1998), penetration (Screiber, 1995) and persistence on foliage (Kudsk et al., 1991). The mode of action of adjuvants is complex (Abbott et al., 1990; Steurbaut, 1993) and in combination with fungicides it may not only result in improved deposition to control pathogens, but also have other effects. Sticker and spreader surfactants may decrease the surface tension of the spray droplets, increasing the bio-availability of the active ingredient. Surfactants may also alter the

cuticle wax components of the targeted area, increasing wettability and spreading (De Ruiter et al., 1990). However, in some instances the effect may be negative, increasing fungal infection, as result of disruption of the cuticle (Marois et al., 1987; Rogiers et al., 2005).

In South Africa, spray technology on viticulture and horticultural crops is a neglected field of research. Specifically, the effective use of adjuvants on grapevines needed to be evaluated. To address these issues, a study was conducted, with the following objectives: (i) to determine the effect of selected adjuvants on spray deposition quantity and quality on grapevine leaves and subsequent biological efficacy of a fungicide in a laboratory study, and (ii) to evaluate selected adjuvants under field conditions and determine the effects of adjuvant concentration and spray volume on deposition.

## **Materials and methods**

### Laboratory studies

#### i) Biological efficacy study

Leaves were sprayed under laboratory conditions to pre-run-off with 1 mL, using a mist sprayer, with a mixture of fenhexamid (Teldor® 500 SC, Bayer) at a recommended dose, a fluorescent pigment (SARDI Fluorescent Pigment, 400 g/L EC; South Australian Research and Development Institute) at 2 mL/L, as well as 15 selected commercial adjuvants, to manipulate the deposition quality of a given quantity of deposited spray. Pigment deposition on leaves was illuminated under black light (UV-A light in the 365 nm region) and visualised under a stereo microscope (Nikon SMZ800) at 10× magnification. Photos of sprayed leaf surfaces were taken with a digital camera (Nikon DMX 1200). Digital images were analysed with Image-Pro Discovery version 6.2 for Windows (Media Cybernetics) software, to determine spray deposition quantity and quality. The sprayed leaves were inoculated with 5 mg dry airborne conidia of *Botrytis cinerea* in a spore settling tower and incubated for 24 h at high relative humidity ( $\geq 93\%$ ). Leaf discs were isolated onto Petri dishes with paraquat-amended water agar and rated 11 days later for development of *B. cinerea* from isolated leaf discs.

#### ii) Histopathology study

Two adjuvants, which had a similar effect in improving spray deposition, but a difference in biological efficacy (as observed in previous laboratory trials), were microscopically investigated on sprayed glasshouse leaves. Leaf inoculation and spray deposition assessments were done as described previously. After 24 and 48 h incubation at a RH of  $> 93\%$ , leaf sections (7 × 5 mm) were stained for 5 minutes in a differential stain. Germination, germ tube lengths and mortality of fungal structures were examined with the aid of a Zeiss Axioskop microscope, equipped with an epifluorescence condenser, a high-pressure mercury lamp, Neofluar objectives and Zeiss filters

02, 06 and 18. With this set-up, protoplast of viable fungal structures fluoresced brilliant yellow-green with filter No. 02, 06 and 18. Protoplasts of dead cells were blue black (Filter 06, 18), whereas cells without protoplast fluoresced white (Filter 02) or yellow (Filter 18).

### iii) Field study

Six adjuvants were tested under field conditions for improvement of foliar spray deposition. The study were divided into four trials, determining the following: (i) optimum volume delivery of the STIHL SR400 motorised backpack mistblower, (to be used in subsequent trials), (ii) evaluation of the surfactants Agral 90, BB5, Break-thru S240, Nu-film-P, Solitaire and Villa51 at the recommended concentrations (iii) effect of varying adjuvant concentration of Agral 90, Nu-film-P, Solitaire and Villa 51 and (iv) effect of different spray volume applications of Agral 90 and Solitaire. Following spray application, exposed leaves were sampled and analysed using the same spray deposition assessment protocol as described previously.

## **Results and discussion**

### i) Efficacy study

The addition of fluorescent pigment to the spray mixture allowed clear visualization of spray deposits on leaves illuminated with black light (Figure 1). Without the addition of an adjuvant, aggregation of pigment particles in remnants of droplets resulted in a distinct droplet effect (Figure 1A). *B. cinerea* incidence on the upper and lower surfaces of the water sprayed leaves (control) averaged 90.4% and 95.8%, respectively. Despite full spray cover of leaves, applications with fenhexamid alone did not completely prevent infection and resulted in 34.6% and 40.8% *B. cinerea* incidence on the upper and lower surfaces of leaves, respectively. With the inclusion of certain adjuvants, the droplet effect was not as distinct as when water or fenhexamid alone was applied. Adjuvants improved deposition quality, resulting in an even spread of pigment particles across the leaf surface (Figure 1C). Wettability of the leaf surface has a major effect on the initial droplet adhesion and retention (Gaskin et al., 2005; Bargel et al., 2006), which could influence the quantity of deposits. Surface-acting-agents (surfactants) present in adjuvants have the potential to lower surface tension (Stevens et al., 1993) of the aqueous solution applied on the target surface, improving droplet wettability and distribution of the active ingredient (De Ruiter et al., 1990; Steurbaut, 1993; Stevens, 1993; Stevens et al., 1993; Gaskin et al., 2005; Bargel et al., 2006). The improvement of spray deposition obtained on leaf surfaces when an adjuvant was included in the fenhexamid mixture, may explain the decrease in *B. cinerea* incidence (2.9-17.1% and 10.0-30.8% on upper and lower leaves, respectively).

### ii) Histopathology study

Epifluorescence microscopy exhibited distinct differences in conidium mortality (31.2% vs. 20.5% vs. 28.1), germination (51.4% vs. 60.4% vs. 57.9%) and germ tube lengths (19.7 vs. 27.8 mm vs. 35.2 mm) for the respective adjuvants Solitaire and Hydrosilicote in combination with fenhexamid and fenhexamid without an adjuvant. Biological efficacy of fenhexamid was improved by Solitaire. This was not the case for Hydrosilicote, even though both adjuvants affected similar deposition quantity and quality (Figure 2). Rogiers et al. (2005) showed that adjuvants may increase susceptibility to infection by *B. cinerea*, counteracting the positive effects of the fungicide. They demonstrated that some adjuvant-fungicide applications could disrupt the epicuticular wax and that the severity of this disruption was dependent on the specific adjuvant.

The laboratory study clearly demonstrated the potential of adjuvants to improve the bio-efficacy of a fungicide directly, through improved deposition on grapevine leaf surfaces; however, bio-efficacy could be influenced by the adjuvant's mode of action.

### iii) Field study

Deposition quantity increased with the increasing spray volumes that were tested (between 27 L/ha to 581 L/ha) using the STIHL SR400 motorized backpack mistblower, but decreased at 698 L/ha, possibly due to run-off. Optimal spray volumes observed ranged from 476 to 698 L/ha. Addition of selected adjuvants at 526 L/ha demonstrated the potential of adjuvants to significantly increase deposition quantity and quality on upper and lower leaf surfaces. Agral 90, BB5, Nu-film-P, and Solitaire significantly improved deposition on upper and lower leaf surfaces compared to a spray with fenhexamid only, as well as the water control. Break-thru S240 and Villa 51 did not improve deposition quantity, although remarkably better deposition quality was obtained.

An adjuvant concentration effect (within the registered concentration range) was evident, particularly those retained on the upper leaf surfaces. Agral 90 and Nu-film-P significantly improved spray deposition at the higher, but not at the lower concentration. Solitaire improved deposition at the lower concentration, while reduced deposition at the higher concentration was attributed to excessive spray run-off. No significant improvement of spray deposition occurred for both concentrations of Villa 51. Higher adjuvant concentration did not necessarily result in better deposition and findings showed that a specific adjuvant may have an optimal concentration at which it can most effectively be used on a specific crop.

Similar to varying adjuvant concentration at a constant spray volume, varying spray volume at a constant concentration also had a significant effect of deposition quantity and quality on grapevine leaves. Spray mixtures with Agral 90 and Solitaire yielded similar deposition values at 500 L/ha compared to fenhexamid only at 720 L/ha, but reduced deposition at the higher spray volume, possibly due to spray run-off of bigger droplets with lower surface tension.

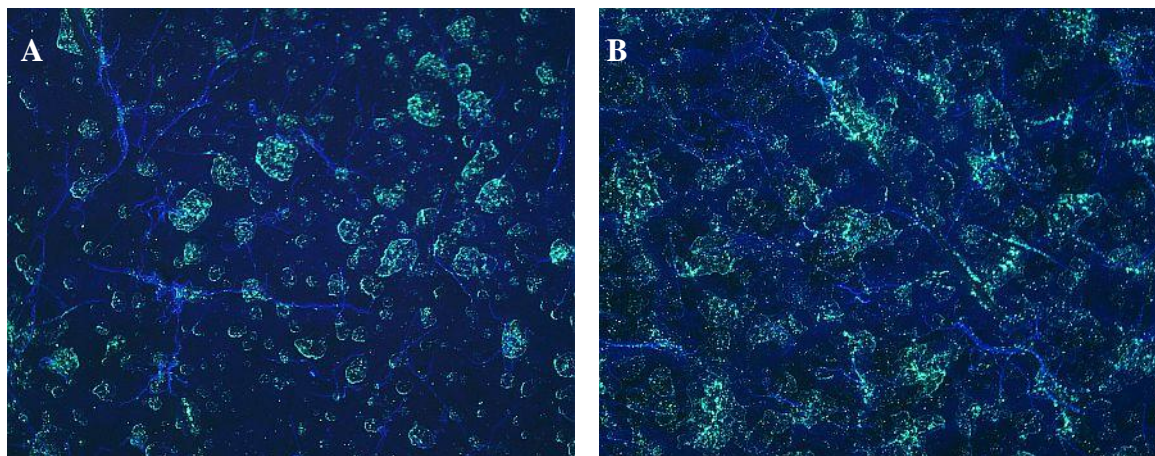
The field study clearly demonstrated the potential of adjuvants to improve deposition quantity and quality, but highlights the necessity to match adjuvant concentration and application volumes on the spray target to achieve maximum spray deposition. A similar observation was made by Gaskin et al. (2002) with a super-spreader adjuvant evaluated at different spray volumes on Chardonnay grapevine foliage. Their results clearly showed decreased droplet retention at a higher spray volume, attributed to more spray run-off. This illustrates the risk of applying adjuvants at wrong concentration at high spray volumes. Therefore, spray volume and adjuvant concentration should be optimised to prevent losses due to run-off. With regard to spray deposition, it is hypothesised that spray volume and adjuvant concentration are inversely correlated, which should be evaluated for each adjuvant product in a specific crop. Gaskin et al. (2002; 2004b) also highlights the need to match adjuvant concentration to application volumes on the spray target to achieve improved spray retention and distribution.

## **Conclusion**

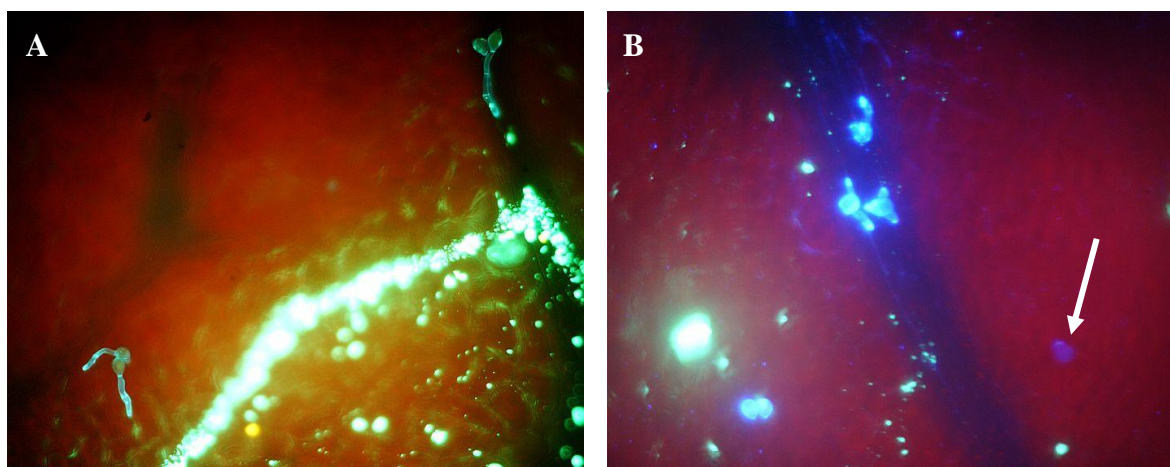
The findings of the laboratory and field study clearly demonstrated the potential, as well as some of the problems, to be encountered when using adjuvants to improve spray application on grapevines. Adjuvants could improve deposition of sprays applied to grapevine leaves, and therewith improve biological efficacy of the applied fungicide against *B. cinerea*. However, the potential deposition of spray droplets is difficult to predict. Adjuvant-fenhexamid combinations, spray volumes and adjuvant concentrations may influence deposition quantity and quality on leaf surfaces. The effect of adjuvants on cuticular wax components on the grape berry could be different to the effect on grapevine leaf surfaces. New horticultural adjuvant technology should be further exploited under field spray programmes to improve spray deposition of agro-chemicals. Unknown aspects, such as high volume application, different agrochemical formulations in the tank mix, applicator technology, canopy size, cultivar type and surface morphology, could affect the spray performance of a horticultural adjuvant. Nevertheless, using an additive to improve spray deposition, the economic benefit of low volume spray application is well recognised as being more cost effective.

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**Figure 1.** Digital images of upper surfaces of Chardonnay leaves sprayed with 1 mL of spray mixture of fenhexamid excluding an adjuvant **(A)**, and a mixture including an adjuvant Agral 90 **(B)** and the SARDI Yellow Fluorescent Pigment, visualised under black light illumination at 10x magnification.



**Figure 2.** Epifluorescence microscope images (400x magnification) of *B. cinerea* on Chardonnay grapevine leaves that were sprayed with a mixture of fluorescent pigment, fenhexamid and selected adjuvants. **(A)** Hydrosilicote + fenhexamid (24 h after inoculation); **(B)** Solitaire + fenhexamid (48 h after inoculation) with dead conidia appearing blue-black under filter set 06, 18 (see arrow).

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