THE EFFECTS OF SILWET L77 ON TRICLOPYR ABSORPTION

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SUMMARY

A nonionic organosilicone surfactant, Silwet L77, enhanced the foliar absorption of triethylamine salt of ^{14}C -triclopyr by the adaxial surfaces of field bean (*Vicia faba* L.) leaves. Fluorescence microscopy of herbicide solutions showed that formulations containing Silwet L77 were able to penetrate rapidly via the stomatal pores. Surface tension of the triclopyr amine solutions was reduced to 22 mN/m. The effects of Silwet L77 on a number of physiological factors, including droplet drying and spreading, were also investigated. A mechanistic model of the uptake process for triclopyr in the presence of Silwet L77 was established.

INTRODUCTION

Silwet L77, an organosilicone surfactant, is a successful adjuvant for a number of woody weed herbicides (Gaskin and Kirkwood 1989) particularly those for gorse (*Ulex europaeus* L.) (Gaskin and Zabkiewicz 1988). Its effect on surface tension and leaf wetting (Goddard and Ananthapadmanabhan 1989) are implicated in the promotion of herbicide uptake by providing an alternative route to cuticular uptake for herbicide entry. This route is via stomatal pores (Field and Bishop 1988; Stevens *et al* 1989) where conditions for diffusion into other leaf compartments could be enhanced. The cuticle lining the substomatal cavity is thinner (Martin and Juniper 1970) thus allowing easier diffusion of the active ingredient into the adjacent cells.

Triclopyr is used for the control of gorse but gorse is a difficult experimental plant and therefore detailed studies on triclopyr performance were carried out on a model plant species, field bean (*Vicia faba* L.). This species was selected on the basis of its similar stomatal characteristics (Buick, in press).

MATERIALS AND METHODS

Field bean seed (cv. Maris Bead) was germinated and potted into 100 mm pots containing potting mix. Plants were raised in a controlled environment cabinet (20-23 °C/15 °C day/night temperature; 70% relative humidity; 14 h photoperiod; PPFD of 300 $\mu\text{moles/m}^2/\text{s}$ and a daily watering regime) for approximately 2 weeks prior to treatment. All experimental procedures were carried out on adaxial leaf surfaces of the third leaves once fully expanded.

Triclopyr triethylamine (IWD 3977 at 4 kg ai/ha in 200 litres/ha) was formulated alone or with the nonionic organosilicone surfactant, Silwet L77 (Union Carbide) at either 0.1, 0.25 or 0.5%v/v. The ^{14}C -triclopyr acid was incorporated into triclopyr triethylamine mixtures to produce ^{14}C -triclopyr triethylamine formulations (Buick *et al* 1989). In two radiotracer studies, each plant received 1 μl as 4 x 0.25 μl droplets, 2 h after the beginning of the photoperiod. Uptake was determined at 1 and 6 h (Experiment 1) or 20, 45, 60 and 120 minutes (Experiment 2) after application by excising the treated leaves, washing (approx. 20 ml) with 70% aqueous methanol and quantifying the radiolabel in the wash by liquid scintillation counting. Uptake was expressed as a percentage of applied radiolabel. There were eight replicates per treatment and the data was arcsine transformed.

Droplet spread was determined for 0.25 μl droplets of the nonlabelled triclopyr triethylamine $\pm$ Silwet L77 formulations by including a fluorescent indicator, Uvitex 2B (Ciba-Geigy) at 1 g/litre in the formulation. Photographs of the dried droplets on leaves were taken under a UV light and image analysed to quantify the area of spread.

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Drying rates were determined as the time taken from 8 x 0.25 µl droplets of a formulation to dry after application to an artificial wax surface. The wax surfaces were made for field bean by a method adapted from Zabkiewicz and Gaskin (1978) (Buick 1990). Droplet drying on clean glass was also determined. Measurements were conducted at 22-23 °C and 60-70% relative humidity. The data for the droplet spread and drying rates were transformed using logarithms.

Formulation absorption was visualised by including the fluorescent indicator, Uvitex 2B, in the triclopyr formulation (1 g/litre) and photographing the patterns of absorbed material under a microscope with a UV excitation source after surface residues were washed off (70% aqueous methanol) at 60 s or 1 h after droplet application. Image analysis was used to quantify the areas of absorbed formulation. A square root transformation of the data was performed.

Surface tension of the triclopyr triethylamine ± Silwet L77 formulations was measured using an adapted droplet-volume technique (Buick *et al* 1989). Twenty replicate measurements were made for each solution and determinations made at 20-22 °C.

RESULTS AND DISCUSSION

After 1 h, the addition of Silwet L77 at all rates increased uptake above the control treatment but this was only significant at 0.25 or 0.5% v/v rates (Table 1). By 6 h, uptake was increased above control but not significantly. The mean absorption areas for each formulation were significantly increased by 0.25 or 0.5% v/v Silwet L77. For each formulation, absorption area was greater after 1 h than after 60 s particularly at 0.25 and 0.5 v/v. Control solutions of herbicide were absorbed predominantly by glandular trichomes, surface lesions and dust while in Silwet L77 formulations, absorption occurred in stomatal pore regions, particularly at 0.25 and 0.5% v/v. From the two sets of experiments, it concluded that the effects of Silwet L77 were most obvious after 1 h.

The fluorescence studies indicated triclopyr entered through stomatal pores when Silwet L77 was present. These results were similar to studies on glyphosate (Field and Bishop 1988) and deoxyglucose (Stevens *et al* 1989). Further evidence for stomatal uptake is necessary and the extent to which overall herbicide uptake is increased because of this alternative route needs elucidation. Possible alternative causes for increased uptake were investigated.

TABLE 1: Uptake (as a percent of applied) of ¹⁴C-triclopyr triethylamine ± Silwet L77 (0, 0.1, 0.25, and 0.5% v/v), absorption area of formulations, surface tension, droplet spread and droplet drying rates of formulations on an artificial field bean wax surface and clean glass. Transformed data and standard errors (SE_x) are given in parentheses:

	Uptake as % of applied		Absorption area (mm ²)		Surface tension (m Nm/m)	Droplet spread (mm ²)	Droplet drying (%/min)	
	1 h	6 h	60s	1 h			Bean wax	Glass
Control	0.8 (4.9)	11.5 (19.6)	0.19 (0.43)	0.20 (0.43)	53.4	49.8 (3.9)	30.5 (3.4)	56.7 (4.0)
0.1% v/v Silwet L77	2.8 (9.5)	12.4 (20.4)	0.10 (0.32)	0.31 (0.56)	22.9	37.0 (3.6)	29.5 (3.4)	56.1 (4.0)
0.25% v/v Silwet L77	6.8 (14.7)	17.1 (24.4)	1.65 (1.19)	7.78 (2.50)	22.4	43.3 (3.7)	31.1 (3.4)	82.2 (4.4)
0.5% v/v Silwet L77	9.7 (18.1)	16.4 (23.8)	2.53 (1.57)	6.73 (2.55)	22.9	67.3 (4.1)	33.6 (3.5)	181.8 (5.2)
SE _x	(1.11)	(1.12)	(0.368)	(0.368)	0.71	(0.55)	(0.03)	(0.07)

The surface tension of all three Silwet L77 formulations were not significantly different and were very low (Table 1), in contrast with the control formulation. If the effect of Silwet L77 on surface tension accounted for improved triclopyr uptake, then the three Silwet L77 triclopyr concentrations should perform equally. As this was not observed, some other mechanism must be involved. According to Schonherr and Bukovac (1972), for stomatal infiltration to occur the surface tension of the formulation must be $\leq$ the critical surface tension value for that surface and solution. An estimate of the critical value for the field bean adaxial leaf surface was found to be 19.5-22.9 mN/m (Buick *et al* 1989). Therefore, the 0.1, 0.25 and 0.5% Silwet L77 solutions in triclopyr amine would be expected to enter stomatal pores. In fact, absorption via stomatal regions was apparent with all three solutions but was only very limited with the 0.1% v/v solution.

The spread of droplets was not significantly different between 0, 0.1 and 0.25% v/v rates of Silwet L77, with the 0.5% v/v treatment being only slightly higher than the other three formulations (Table 1). Since maximum uptake was achieved at both the 0.25 and 0.5 % v/v rates of Silwet L77, the greater droplet spread at 0.5% v/v was unlikely to have contributed much to uptake.

As the rate of Silwet L77 increased, droplet drying increased, particularly on the clean glass surface (Table 1). On field bean wax, there was not a significant increase in drying rate. Complete wetting occurred with the Silwet L77 formulations on glass but not on the field bean wax. Dissolution of the field bean wax surface beneath the droplets took place with all four triclopyr formulations, probably reflecting the action of the active ingredient. The rapid droplet drying rates observed at high surfactant concentration were expected because of the greater droplet spread. The increase in droplet spread on field bean leaves was not dramatic which is in accordance with the non-significant change in drying rate on the extracted field bean wax. Maintaining the solute in solution tends to promote diffusion of the active ingredient into the plant (Price 1982). Silwet L77 significantly increased the rate of droplet drying when used at 0.25 or 0.5% v/v which is the converse of that required to promote uptake. Therefore, Silwet L77 does not appear to affect droplet spread or drying rate in ways which would significantly promote uptake.

A second radiotracer study monitored the triclopyr triethylamine uptake over time. The 0.25% v/v rate of Silwet 77 was selected since evidence suggested that it was able to enhance uptake to the same degree as the 0.5% v/v rate without any negative effects on translocation processes (Buick 1990).

Uptake was significantly increased by 0.25% v/v Silwet L77 (Table 2); the pattern of triclopyr uptake showed an initially rapid uptake rate which declined between 20 and 45 minutes. The first phase probably represents an initial mass flow of material into the substomatal cavities. By 20 minutes, this flux began to slow as substomatal regions became saturated. The second phase occurred after 45-60 minutes when uptake began to increase again with the rate approximately equal to control after 45 minutes. It is

TABLE 2: Uptake (as a percent of applied) of ^{14}C -triclopyr triethylamine with and without added Silwet L77 (0.25% v/v) at 0, 20, 45, 60 and 120 minutes after application of 0.25 μl droplets of formulation to the field bean adaxial leaf surface. Arcsine transformed data and standard errors (SE_x) are given in parentheses.

Time (minutes)	Uptake of triclopyr as % of applied	
	Control	Silwet L77 (0.25% v/v)
0	0	0
20	1.3 (6.3)	6.0 (14.0)
45	1.3 (6.3)	6.4 (14.3)
60	3.1 (9.8)	10.9 (19.0)
120	8.8 (17.0)	17.5 (24.6)
SE_x	(1.16)	(1.16)

believed that by 45-60 minutes, the triclopyr active ingredient had perturbed the surface wax and cell layers to permit a more rapid diffusion of active ingredient into the plant. This was the same for both the control and Silwet L77 treatments. Apparent dissolution of the field bean wax surfaces with both Silwet L77 and non-Silwet L77 mixtures suggested a reaction between triclopyr and components of the field bean wax. Scanning electron microscopy supported the dissolution or breaking down of surface layers and/or cells on the field bean leaf, which first became apparent 30-60 minutes after triclopyr formulations had been deposited on the leaf (Buick 1990).

The initial difference in uptake between the control and Silwet L77 treatments was established in the first 20-45 minutes with Silwet L77 having a significantly greater uptake. A two phase model of triclopyr uptake over time in the presence of Silwet L77 is proposed for the field bean — triclopyr — Silwet L77 system. In phase 1, herbicide uptake was dramatically enhanced by the mass flow of solution through stomatal pores, which did not take place with the control formulation. The second phase began approximately 45-60 minutes after application by which time the external barriers to herbicide diffusion had become sufficiently reduced to permit a relatively rapid movement of active ingredient into the plant and was the same for both control and Silwet L77 formulations. Stomatal uptake appears to be the primary mode of action for Silwet L77 in promoting uptake of triclopyr.

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