**POSTHARVEST CONTROL OF DIAPAUSING TWO-SPOTTED
SPIDER MITE *TETRANYCHUS URTICAE* KOCH ON FRUIT.
II EFFICACY OF PULSE ON APPLES**

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SUMMARY

Experiments were carried out to determine the effect of Pulse, a spray adjuvant, combined with 100 ppm chlorine, on the removal and mortality of two-spotted spider mite *Tetranychus urticae* (TSM) on apples. More than 96% of diapausing TSM which had not spun webs were removed from apples when dipped in water or Pulse. There was no relationship between Pulse concentration and percentage of mites removed. Of those mites remaining on the apples, all were killed in concentrations of 0.75% (v/v) Pulse and above. Thirty-nine to 65% of diapausing TSM were removed when apples on which webbing was established were dipped in 1.25% (v/v) Pulse, and complete kill of the remaining mites was achieved.

Keywords: two-spotted spider mites, Pulse, postharvest dip, removal, mortality

INTRODUCTION

In the apple orchard, two-spotted spider mites (*Tetranychus urticae*; TSM) are found on leaves, branches and fruit, where they produce webbing. This webbing plays an important part in protecting mites from predators, and is also used for dispersal, in mating behaviour and in protection from climatic factors (Gutierrez and Helle 1985). Webbing may also protect the diapausing TSM female from pesticide particles (Gerson 1985).

Dentener and Peetz (1992) compared insecticidal soaps and spray adjuvants for their effects on diapausing TSM mortality using a slide dip method. They found that Pulse was the most toxic of the products tested, providing 99% mortality of diapausing TSM at a concentration of 0.48% (v/v) in water containing 100 ppm chlorine. Similar results were described for the gorse mite by Searle *et al* (1990).

In this paper we describe experiments to determine the effects of Pulse, combined with 100 ppm chlorine, on diapausing TSM on apples. Apples were used as a substrate because diapausing TSM infest apple fruit in late summer and can be a problem for exporters. TSM were caged on apples and exposed to different concentrations of Pulse plus chlorine, to provide dose-mortality data. Separate experiments were carried out to determine if mites who had spun webbing on apples could be washed off with Pulse.

MATERIALS AND METHODS

Apples cv. 'Granny Smith' were obtained from the DSIR Research Orchard in Kumeu. The fruit was kept insecticide-free for at least 8 weeks prior to harvest, but otherwise received a normal spray programme.

Diapausing TSM were collected from bean plants on cardboard traps, and stored for about 5 weeks at 4°C in darkness before use in these experiments (Dentener and Peetz 1992).

Mites caged on apples

Between 40 and 60 TSM were transferred with a fine (000) camel hair paint brush from cardboard traps to cages made of flexible semi-oval PVC rings, measuring ca. 20 x 30 mm, and 15 mm high (internal volume 5-6 ml). One side of the ring was sealed with fine metal gauze (64 squares per mm²), and each cage was connected to

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one individual apple with a thin strip of Bostik Blu-tack®. Mites were left in the cages at ambient temperature (ca. 20°C) for about 3 hours before treatment. Treatments were administered by filling cages with a range of five concentrations of Pulse containing 100 ppm chlorine in the solution, using a pipette. Five cages were used per concentration of Pulse and the solution was left in the cage for about 10 seconds. The cages were then allowed to drain for 15 minutes at ambient (ca. 20°C) temperature and placed overnight (18 hours) at 20°C, L:D 16:8 before assessment. Two controls were used. In the first, fruit was either dipped in (or cages filled with) water containing 100 ppm chlorine, and in the second the fruit was kept dry ("untreated") before being held overnight in the same conditions as the water-treated fruit. The experiment was carried out three times.

Mites on apples without webbing

Forty mites were transferred onto an area of each apple which had been degreased with 70% ethanol before a circular insect barrier of Fluon® (poly-tetra-fluoro-ethylene dispersion agent) (ca. 50 mm in diameter) was painted on the cheek. Within 10 minutes of inoculation with TSM, five apples per treatment were submerged for 3 minutes in a range of five Pulse concentrations (0.5% - 1.5% v/v), left for at least 15 minutes at ambient (ca. 20°C) temperature and placed overnight (18 hours) at 20°C, L:D 16:8 before assessment. The 3 minute immersion period was based on the minimum time fruit could spend in a commercial water dump prior to packing (H. Tough, pers. comm. 1991). Two control treatments (two apples in each) were conducted as described in the previous experiment.

Mites on apples with webbing

Sixty mites were transferred into each cage which was connected, as described above, to either the calyx or stem end of the fruit. They were kept at 20°C for 42 hours to allow the mites to spin webbing. The cages were removed before dipping, and any mites on webbing remaining in the cage were carefully transferred onto the apple. Four apples with mites at the calyx, and four apples with mites at the stem end of the fruit were submerged for 3 minutes in Pulse at a concentration of 1.25% v/v. The choice of concentration was based on the high mortality of TSM obtained in previous experiments. In addition, two apples with mites at the calyx, and two apples with mites at the stem end of the fruit were submerged for 3 minutes in water as "controls". After dipping, all apples were left for at least 15 minutes at ambient (ca. 20°C) temperature and placed overnight (18 hours) at 20°C, L:D 16:8 before assessment.

In all of the above experiments, the survival of the mites was assessed under a binocular microscope (x10-15). They were recorded as "alive" if leg or body movement was detected when gently prodded with a fine (000) camel hair paint brush. Dose-mortality data were analyzed using probit analysis, based on logarithm of the concentrations, to derive LC_{50} and LC_{99} estimates (i.e. the estimated concentrations to achieve 50 and 99% mortality respectively) (Finney 1971).

RESULTS AND DISCUSSION

Mites caged on apples

Results from the individual apples in each replicate were grouped together since there were no statistical differences between the data for individual fruit.

No mortality was observed in the "untreated" control, and mortality in the "water" controls varied between 0 and 1.3%. Mean LC_{50} values, mean LC_{99} values and 95% confidence intervals were calculated, based on three separate replications, on a logarithmic scale and transformed back (Table 1). The mean LC_{99} of 1.84% for TSM on apples is much higher than that found by Dentener and Peetz (1992) for diapausing TSM dipped in a solution of Pulse containing 100 ppm chlorine (LC_{99} : 0.48%), using a slide dip method. The variability between replicates makes this method unsuitable for determination of dose-mortality data for TSM when using a postharvest dip, and further experiments described in this paper were carried out with TSM inoculated on apples, and dipped in solution.

TABLE 1: Estimated LC₅₀ and LC₉₉ values for diapausing TSM held in cages on apples and flooded with a range of concentrations of Pulse containing 100 ppm chlorine. 95% fiducial limits in brackets.

Replicate	% v/v Pulse			
	LC ₅₀		LC ₉₉	
1	0.02	— ^a	1.91	— ^a
2	0.07	— ^a	2.49	— ^a
3	0.28	— ^a	1.30	— ^a
MEAN	0.07	(0.003 - 1.95)	1.84	(0.82 - 4.13)

^a Meaningful confidence intervals could not be calculated

TABLE 2: The effect of Pulse and water (both containing 100 ppm chlorine) on “washing-off” and mortality of diapausing TSM on apples. Mites were inoculated on the cheek of the apple (no webbing), or the calyx or stem end (webbing), and dipped in the solution for 3 minutes.

Experiment	Treatment	Number (%) of mites		
		No. removed (%)	No. remaining on apple	No. dead on apple (%) mortality
No webbing ^a	Untreated	63 (78.7)	17	0 (0.0) ^c
	Water	77 (96.2)	3	2 (66.7) ^c
	0.5% Pulse	193 (96.5)	7	6 (85.7) ^c
	0.75% Pulse	198 (99.0)	2	2 (100.0) ^c
	1.25% Pulse	198 (99.0)	2	2 (100.0) ^c
	1.5% Pulse	194 (97.0)	6	6 (100.0) ^c
Webbing ^b	Water (calyx)	29 (48.3)	31	1 (3.2)
	Water (stem)	15 (25.0)	45	1 (2.2)
	1.25% Pulse (calyx)	47 (39.2)	73	73 (100.0)
	1.25% Pulse (stem)	78 (65.0)	42	42 (100.0)

^a Inoculated 200 (each concentration) and 80 (water and untreated each) mites on cheek of apple

^b Inoculated 120 mites (1.25% v/v Pulse) on 4 apples at calyx and 4 apples at stem end; 60 mites (water) on 2 apples at calyx and 2 apples at stem end of fruit

^c Number of dead mites recovered too low for analysis.

Mites on apples without webbing

Only about 21% of the mites inoculated on the “untreated” apples were recovered. About 96% of the mites were washed off the apples when they were dipped in water containing 100 ppm chlorine for 3 minutes (Table 2: no webbing). Similarly, between 96.5% and 99% of the mites were washed off the apples during dipping in all concentrations of Pulse indicating that there was no relationship between Pulse concentration and percentage TSM removed. Comparison of removal of mites in the “untreated”, “water control” containing 100 ppm chlorine, and Pulse showed statistical differences ($P=0.025$) between the treatments. However, the absolute differences were so small that the differences in treatment efficacy are of no practical consequence. The number of mites remaining on the fruit was too low for analysis. Of those mites remaining on the apples, all mites were killed when exposed to concentrations of 0.75%

(v/v) Pulse and higher. This concentration is much lower than the mean LC₉₉ of 1.84% found for the caged mites, and similar to results described by Dentener and Peetz (1992).

Mites on apples with webbing

From those apples on which mites had been allowed to establish webbing, about 48% (calyx end) and 25% (stem end) of the mites were removed when dipped in water (Table 2: webbing). The mortality of mites remaining on the apples after a water dip varied between 2% (stem end) and 3% (calyx end). However, no mites survived on apples dipped in a 1.25% (v/v) Pulse solution. About 39% (calyx end) and 65% (stem end) of the inoculated mites were "washed-off" by this dip.

The percentage of mites removed from apples dipped in water or Pulse was much lower (25-65%) when the mites were allowed to spin webbing at the calyx or stem end of the fruit, than when they were on the cheek and no webbing was present (about 97-99%) (Table 2). The high loss of mites (78.7%) from the cheek of apples in the undisturbed "untreated" apples suggests a high level of mobility of the mites in this treatment.

Of the mites which were inoculated at the calyx or stem end, and which were allowed to establish webbing, more were recovered alive from the calyx end than from the stem end after dipping in water containing 100 ppm chlorine (Table 3). Mites which had not established webbing and were dipped in Pulse at concentrations of 0.75% v/v and above were all dead or washed off (Table 2). Mites which had established webbing and were dipped in 1.25% (v/v) Pulse, were all found dead in the position where they had been inoculated. This indicates that webbing inhibits removal of TSM, and that treatment with Pulse at 1.25% (v/v) results in complete TSM mortality.

TABLE 3: Positions on apple from which mites, which had spun webbing, were recovered in washing-off experiments when inoculated at calyx or stem end of the fruit. For absolute numbers, see Table 2.

Inoculation position	Treatment	% of total mites recovered					
		Live mites			Dead mites		
		Cheek	Calyx	Stem	Cheek	Calyx	Stem
Calyx	Water	0	71.0	25.8	0	3.2	0
Stem	Water	0	57.8	40.0	0	0	2.2
Calyx	1.25% Pulse	0	0	0	0	100	0
Stem	1.25% Pulse	0	0	0	0	0	100

CONCLUSIONS

Dentener and Peetz (1992) showed that Pulse was effective in killing diapausing TSM. Pulse at 0.48% (v/v) containing 100 ppm chlorine caused 99% mortality. Research described in this paper has shown that 39-65% of diapausing TSM (which had spun webs) were removed from the calyx or stem end of apples by dipping in 1.25% Pulse solution containing 100 ppm chlorine, and that complete kill of the remaining mites was achieved with this treatment. By contrast, 100 ppm chlorine in water with or without Pulse removed 96% of TSM from the cheeks of apples when the mites had not spun webbing. The few remaining mites were killed by concentrations of 0.75% Pulse and higher.

Pulse shows potential as a postharvest mite disinfestation dip since it results in high mortality of TSM. Pulse residues on fruit may produce undesirable flavours. Further research is required to determine the effect of washing Pulse off the produce following dipping, the resulting effect on mite mortality, the efficacy of these procedures on a commercial scale, and its effect on fruit flavour.

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