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# EFFECTS OF PESTICIDE DEPOSIT PATTERN ON RATE OF CONTACT AND MORTALITY OF *APHYTIS HOLOXANTHUS* (HYMENOPTERA: APHELINIDAE)

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#### ABSTRACT

A laboratory study was designed to determine the pesticide droplets deposition on leaf surfaces and its effect on parasite behavior and mortality. The rate of parasite/deposit contact increased as the number of deposits increased, even though total volume of deposit remained constant. The bioassay results indicated that mean percent mortality of *Aphytis holoxanthus* in the carbaryl treatment was highest on leaf discs with smaller and more abundant deposits than larger and fewer deposits.

Key Words: Florida red scale,  $Aphytis\ holoxanthus,$  deposit pattern, contact rate, parasite

#### RESUMEN

Se realizó un estudio de laboratorio para determinar el patrón de deposición de gotas de plaguicida en la superficie de las hojas y su efecto en la tasa de contacto y la mortalidad de *Aphytis holoxanthus*. La tasa de contacto de parásito/depósito aumentó a medida que el número de depósitos se incrementó, aún cuando el volumen total de los depósitos se mantuvo constante. El porcentaje de mortalidad promedio de *Aphytis holoxanthus* en el tratamiento con carbaryl fue más alto en los discos de hojas con depósitos más pequeños y abundantes que en los discos con depósitos más grandes y escasos.

Biological control of Florida red scale (FRS), *Chrysomphalus aonidum* (L.) (Homoptera: Diaspididae), by the introduction of *Aphytis holoxanthus* DeBach has permanently reduced the pest status of Florida red scale to the extent that citrus growers generally have forgotten that during the 1950s it was one of their most serious pests (Browning, 1994). This parasite searches actively on citrus foliage and fruit to find its armored scale host and can be negatively affected by pesticide deposits due to high rates of contact associated with its walking on plant surfaces (Rosen, 1973). The impact of pesticide deposits on parasite survival may be dependent upon the pattern of deposition, i.e. size and number of deposits and on the pesticide concentration of the deposit.

Different spray techniques are used to achieve spray deposition efficiency on citrus leaves to maximize control of target pests, but improvements in deposition may also severely disrupt host-parasite relationships. By measuring quickly and accurately the droplet size of released sprays, the effects of droplet size on deposit and on biological activity of the spray in relation to a specific crop pest can be more clearly defined (Akesson & Yates, 1989). The spray deposition efficiency of orchard airblast sprayers generally is considered to be low and is influenced by droplet size (Reichard et al., 1977). New leaves have less surface waxes and showed slightly better deposition pattern than mature waxy leaves (Salyani, 1988). The bottom surface of leaves, having a more rough texture, captured more droplets than the top surfaces (Salyani, 1988). This study was designed to determine the pesticide deposition on leaf surfaces and its effect on parasite behavior and mortality. The objectives of this study were to characterize the effects of different pesticide deposit patterns on rates of parasite contact during leaf searching, and to determine the impact of contact rate on adult parasite mortality.

# MATERIALS AND METHODS

Adult parasites were allowed to search on leaf surfaces containing various patterns of pesticide deposits. The total amount of pesticide and carrier delivered was the same among all patterns. Parasite behavior was evaluated during searching, and rates of contact with pesticide deposits were calculated for each deposit pattern.

Bioassays were conducted according to Rehman et al. (1998), using each deposit pattern to determine the influence of pattern on parasite mortality induced by the pesticide residues. These experiments were conducted under laboratory conditions of  $24 \pm 2^{\circ}$ C and 60% R.H.

# A. Holoxanthus Behavior on Different Deposit Patterns

Four deposit patterns of carbaryl and untreated controls were replicated three times. The pesticide treatment was carbaryl wettable powder (Sevin 80S: Rhone-Poulenc, Research Triangle Park, NC 27709) at 1.5 g carbaryl per liter of water (1.25 lb per 100 gal). Water served as the control. Kaolin clay (500 mg) was added to all solutions to improve the visibility of deposits. Deposit patterns of 1, 2, 5 and 10µl droplets were made with a 10 µl ±1% micro dispenser (Drummond, USA). The leaf deposits produced by these volumes are found in field applied deposits (Salyani and Fox, 1994). A total of 20 µl of each solution was dispensed onto  $2 \times 1.7$  cm 'Hamlin' orange rectangular leaf patches. The deposits (20 droplets, 1 µl; 10 droplets, 2 µl; 4 droplets, 5 µl; or 2 droplets, 10 µl) in each replication were air dried for 90 minutes. The actual deposit size for each pattern was obtained by measuring the deposit through a microscope. The length and width of the four deposits for each pattern were measured and the diameter of each deposit was recorded in square millimeters (mm<sup>2</sup>).

Twenty adult *Aphytis holoxanthus* were exposed to each deposit patterns in each replication by confining them in a small ventilated petri dish for each single leaf patch. Each replication was recorded for 2 h by time lapse videography at 60 frames/ sec (Panasonic, Osaka, Japan). Observations were then interpreted and recorded during the first 10 min, the middle 10 min (from 55-65 min), and the final 10 min of the 2 h exposure period. In each case, the observations were made by analyzing the first 10 parasite searching events on each leaf patch. One searching event consisted of an individual adult parasite traversing the leaf patch. The total time on the patch and number of encounters with spray deposits were recorded during review of the video-taped exposures. Rates of contact were then calculated by dividing the number of contacts between the parasite and a deposit droplet, by the total time the parasite spent on the leaf patch. These data were analyzed by PROC GLM, comparing the rates of contact for different deposit patterns during the three time intervals monitored (first 10 min, mid 10 min, and final 10 min) and the LSD procedure (SAS Institute, 1988).

### A. holoxanthus Mortality

The four deposit patterns described above for each treatment of carbaryl and control were repeated on  $4.5 \text{ cm}^2$  leaf discs punched from field collected leaves and were

air dried for 90 minutes. Four replicates of each treatment were placed into ventilated plastic 4.9 cm<sup>2</sup> diameter petri dishes. The bioassays were conducted as described by Rehman et al. (1998). After 24 h of exposure of 20 adult *A. holoxanthus*, mortality data were collected from each deposit droplet pattern by detaching the petri dishes from the ventilation system and counting the live and dead parasites in each petri dish. The mean percent mortality was calculated and compared between deposit pattern and presence or absence of carbaryl. The data were analyzed with ANOVA and Fisher's least significant difference (LSD) test (SAS Institute, 1988). Abbott's corretion for control mortality was not used because control mortality was below 5%.

#### RESULTS

#### A. holoxanthus Contact Rate

As the number of droplets in the deposit pattern increased, the rate of parasite/deposit contact increased in the controls, even though total volume of deposit remained constant (Table 1). In the carbaryl treatment increased contact occurred with 20 droplets at all times compared to the other droplet treatments (Table 1). There were essentially no differences between the contact rate of other droplet numbers (Table 1). Also, the rate of contact was highest on leaf discs with 20 deposits in the control and treatment. Although the size of individual deposits increased with fewer droplets, the rate of contact with larger deposits was lower than with the more numerous smaller deposits. The droplet sizes and droplet volume for different deposit droplet numbers are given in Table 1. According to PROC GLM (SAS Institute, 1988), the mean rate of contact to different numbers of deposits was highly significant (F = 13.16; df = 11; P < 0.0001 and  $r^2$  = .92), being directly proportional to the number of deposit droplets, particularly in the control treatment. Observations of different time intervals during the 2 h exposure indicated that the rate of contact within the different 10 min intervals for pesticide and non-pesticide treatments was also significant (F = 6.77, df = 2, P < .0107). Rates of contact for patches with 20 deposits of carbaryl were higher during the first 10 min and decreased afterward, in comparison to the control, where rates of contact increased during the middle and final 10 min (Table 1). Carbaryl treatments with 10 and 20 deposit droplets showed evidence of parasite effects during the middle 10 min and final 10 min periods. Adult parasites became sluggish and their mobility decreased. Few sluggish or immobile parasites were observed on leaf patches with 2 or 4 deposits of carbaryl. Control treatments showed no evidence of negative effects on parasite activity during the 2 h exposure period. The parasites spent most of their time walking or grooming on the leaf surface, but responded to detection of deposits on the leaf surface by exhibiting antennation and probing behavior. Occasionally, parasites were observed to stop on droplet deposits, circle around them and tap or probe the margins of the deposits with their ovipositers. Contact with deposits appeared to be random. Parasites did not appear to be either attracted or repelled by the deposits in either the carbaryl or control treatments.

# A. holoxanthus Mortality

The bioassay indicated that mean percent mortality of *A. holoxanthus* in the carbaryl treatment was highest on leaf discs with the smallest and more abundant deposits. The mean percent mortality on leaf discs with 20 and 10 deposits was 53.8% and 45.2%, respectively. There was no mortality on leaf discs with 4 deposits of carbaryl; however, some parasites were sluggish and a few were immobile. Parasite mor-

Treatments				$\overline{x}~$ rate of contact (# of contact/sec) (± SE)		
	$Deposit \ numbers \qquad Drop \ vol \ (\mu l)$		$\begin{array}{c} Deposit\ diameter\\ (mm^2)\ (\pm\ SE) \end{array}$	First 10 minute	Mid 10 Minute	Final 10 Minute
control	2	10	5.6 (1.3)	$0.07^{a}(.04)$	$0.23^{a}$ (.08)	$0.32^{a}(.06)$
	4	5	5.1(2.0)	$0.19^{\rm b}(.06)$	$0.49^{\rm b}(.08)$	$0.59^{\rm b}(.05)$
	10	2	4.3 (1.2)	$0.41^{\circ}(.04)$	$1.01^{\circ}(.18)$	$0.68^{\circ}(.09)$
	20	1	3.8(0.5)	$0.75^{d}$ (.13)	$1.31^{d}(.39)$	$1.49^{d}$ (.28)
			Mean	0.36	0.76	0.74
carbaryl	2	10	6.0 (.8)	$0.11^{a}(.03)$	$0.29^{a}(.16)$	$0.09^{a}$ (.03)
	4	5	5.4(0.8)	$0.16^{a}(.09)$	$0.39^{a}(.14)$	$0.15^{*}(.02)$
	10	2	4.4(0.7)	$0.29^{\rm b}(.07)$	$0.29^{a}(.19)$	$0.25^{\text{ab}}(.09)$
	20	1	3.9 (0.4)	1.07 c (.28)	$0.69^{\text{b}}(.11)$	$0.67^{\circ}(.13)$
			Mean	0.48	0.42	0.29
			Grand mean	$0.42^{\text{a}}$	$0.59^{ m b}$	$0.52^{ m b}$

TABLE 1. MEAN RATE OF CONTACT OF APHYTIS HOLOXANTHUS TO DIFFERENT DEPOSIT PATTERNS.

SE = Standard error of mean

Means followed by a different letter for each treatment in a column and column grand means followed by a different letter in each row are significantly different at p < 0.05 according to LSD multiple range test.

tality in deposit patterns without carbaryl (control) averaged 3.7% with 2 deposits and 1.2% with 10 deposits (Fig. 1) indicating no significant effect of deposit size or number.

# DISCUSSION

According to Himel (1969), as the diameter of the average droplet size decreases, the total number of spray droplets increases markedly. This increases the potential for contact of the droplets with target insects. In another study Himel and Moore (1969) mentioned that most commercial spray devices produce spray droplets whose size (diameter) ranges from a few microns to hundreds of microns. The efficiency of any pesticide spray and the degree of control obtained is a function of the ultimate point of deposition of each of the pesticide spray droplets produced. In this study A.



Fig. 1. Mean % mortality of adult *Aphytis holoxanthus* after 24 h exposure to treated leaves with different numbers of deposit sizes. Error bars = standard error of the mean.

*holoxanthus* did not react differentially to the carbaryl and control deposit patterns. Random contact of *A. holoxanthus* with deposits in both the carbaryl and control treatment suggested that there was no obvious attraction or repellency by these deposits. The lower rates of contact in the pesticide treatments of 20 deposits at the middle 10 min and final 10 min periods of the 2 h exposure were due to sluggish parasite behavior. There was no obvious trend for other deposit sizes. In contrast, the parasites on control deposits were very active throughout the 2 h exposure period.

Salyani and McCoy (1989) concluded that decreasing spray droplet size increased citrus rust mite control. Our study showed that, for a constant amount of pesticide applied, increased numbers of smaller droplet deposits resulted in increased contacts between actively searching *A. holoxanthus* adults and static deposits. Greater mortality was experienced within treatments in which rates of contact were highest. Thus, improved coverage of foliar pesticide applications may magnify the mortality rate of parasites.

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