Effects of a Kaolin-Based Particle Film on Obliquebanded Leafroller (Lepidoptera: Tortricidae)

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ABSTRACT Studies were conducted in 1997 to evaluate the effects of the kaolin-based particle film formulation M96–018 on adults, eggs, and larvae of the obliquebanded leafroller, *Choristoneura rosaceana* (Harris). Particle film treatments significantly reduced female longevity, mating success, and number of egg masses oviposited compared with moths on untreated apple leaves in sleeve-cage and screen-cage tests. No differences in mating success or oviposition were caused by the application rates and coverage density of M96–018 on foliage. Females avoided ovipositing on particle film-treated leaves in choice tests. Larval hatch was not affected by topical application or residual exposure to M96–018. Larval weight gain and pupal weight were significantly reduced and larval mortality increased in no-choice feeding tests with M96–018. In choice tests, larvae preferred to feed on untreated leaf surfaces. The negative effects on larval development and survivorship on M96–018-treated foliage did not differ across a fourfold difference in spray application rate. A significant reduction in the number of infested shoots was found in orchard trials when M96–018 was applied before bud break in late March compared with untreated trees. No reductions in larval densities were found compared with an untreated control following prebloom and postbloom applications.

KEY WORDS *Choristoneura rosaceana*, apple, pest management, particle film, kaolin

THE OBLIQUEBANDED LEAFROLLER, *Choristoneura rosaceana* (Harris), has become a key pest in tree fruit orchards throughout North America (Reissig 1978, Madsen et al. 1984, AliNiazee 1986, Barnett et al. 1991, Beers et al. 1993, Long et al. 1997). Similar to most other orchard tortricid pests, *C. rosaceana* larvae feed primarily on foliage but they also damage fruit, especially when the fruit and foliage are in proximity or the fruit is in clusters (Beers et al. 1993). Control measures for *C. rosaceana* have included a number of organophosphate insecticides (Brunner et al. 1994); however, concerns about the health risks of pesticide residues on fruit and the passage of the Food Quality Protection Act in 1996 could eliminate or seriously restrict the continued use of this class of chemical in the United States. In addition, *C. rosaceana* populations have developed resistance to organophosphate insecticides (Lawson et al. 1997). Alternatively, the use of microbial insecticides such as *Bacillus thuringiensis* Berliner have increased in recent years, but these materials are shortlived and require multiple applications to be effective (Knight 1997). The use of sex pheromones for mating disruption of *C. rosaceana* has been investigated, but it has not been widely adopted by growers (Knight et al. 1998). Clearly, new tactics are needed to manage *C. rosaceana.*

A particle film technology has been proposed as a new paradigm in pest management (Glenn et al. 1999). Crops are sprayed with a hydrophobic surface-coated kaolin clay particle that creates a protective barrier against both plant pathogens and plant-feeding arthropods. Kaolin is a white, nonabrasive, inert aluminosilicate mineral that is widely used in a variety of industrial applications, including paints, cosmetics, and pharmaceuticals. However, experimental trials with untreated kaolin clays in pest management historically have been only moderately effective (Alexander et al. 1944, David and Gardniner 1950). Recently, USDA–ARS and the Engelhard (Iselin, NJ) formed a partnership to develop coated kaolin-based particle films for use in agriculture. Preliminary studies evaluated the effect of treating apple and pear foliage with the M96–018 formulation against pear psylla, *Cacopsylla pyricola* (Förster); spirea aphid, *Aphis spireacola* Potch; twospotted spider mite, *Tetranychus urticae* Koch; and the potato leafhopper, *Empoasca fabae* Harris (Glenn et al. 1999). Densities of these pests were significantly reduced on treated versus untreated foliage in greenhouse and field trials. The effects appeared to be caused by repellency, disruption of feeding and oviposition, and increased mortality (Glenn et al. 1999). Similar studies against the major lepidopteran pests of tree fruits, such as codling moth, *Cydia pomonella* L., and tortricid leafrollers have not been reported. The objective of our study was to evaluate the activity of the M96–018 formulation against the adults, eggs, and larvae of the tortricid leafroller, *C. rosaceana.*

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Materials and Methods

Laboratory Rearing of *C. rosaceana*. All studies were conducted with eggs, larvae, and adults obtained from a laboratory colony of *C. rosaceana* originally collected in 1992 from several apple orchards in Grant County, WA. Larvae were reared on a synthetic pinto bean diet (Shorey and Hale 1965) at 24°C and a photoperiod of 16:8 (L:D) h in 30-ml plastic cups. Adults were supplied with a cotton wick saturated with a 10% honey solution. Egg masses were laid on wax paper sheets. Only virgin moths 2–3 d old were used in the various adult tests.

Particle Film Application. The standard rate of M96–018 used in all tests was 0.03 kg/l plus 0.04 liters of methanol per liter of water. M96–018 was mixed separately with methanol and then added to water in the sprayer tank. Continuous agitation was required to keep the material in solution. The effect of spray rate against adults and larvae was evaluated in two studies. M96–018 was applied in these tests at three rates: 0.06, 0.03, or 0.015 kg/l water; the concentration of methanol was kept unchanged. M96–018 was applied in the field with either a nonpressurized backpack sprayer (Solo Port 423, Postfach, Germany) or an engine-driven, pressurized 95-liter sprayer (Rears Manufacturing, Eugene, OR) equipped with a spray gun (Gunjet No. 2; Teejet, Boise, ID) running at 7.0 kg/cm^2. Seedlings in the greenhouse were sprayed with a hand-pumped 2.0-liter sprayer. Trees and seedlings were sprayed until drip and were visually inspected and resprayed to ensure complete coverage. Field studies were conducted during 1997 either in a 20-yr-old 'Golden Delicious' or a 5-yr-old 'Fuji' orchard at the USDA experimental farm (Moxee, WA) or in a nonbearing 3-yr-old Fuji orchard near Brewster, WA.

The residual concentration of M96–018 on foliage was measured with a methanol leaf extraction procedure. One fully expanded leaf from the midbranch of the current season's wood was removed from each of five branches of a tree, placed in a plastic bag, and refrigerated at 2°C until used (<2 wk). Three trees from each treatment were sampled on each date. Leaves were individually washed in 25 ml of methanol in the bottom of a 9-cm glass petri plate with a short bristled, 1-cm-wide paint brush to agitate the kaolin free of the leaves. Methanol was replenished as needed to clean all five leaves in the same plate. The methanol was allowed to evaporate for 48 h in a fume hood. Plates were weighed, carefully cleaned and dried, and reweighed. Kaolin particle weights were derived by subtraction. Leaf area was measured with a LiCor 3000 area meter (LiCor, Lincoln, NE), and residue per square centimeter was obtained by dividing the residue weight by twice the measured surface area. Residue was corrected to that caused by particle films alone by subtraction of corresponding values from untreated control trees. Reported are means of the three replicate trees.

Residual Effects of M96–018 Against Adults. The effects on mating and larval hatch of *C. rosaceana* were studied in the Golden Delicious orchard. Ten replicated trees were treated with four applications of the standard rate of M96–018 (0.03 kg/l) on 13 May, 2 and 16 June, and 11 July. One male:female pair was placed in sleeve cages (12 by 12 by 36 cm) on each M96–018-treated and on 10 untreated trees on 3 and 22 July. Sleeve cages were removed after 10-14 d, and the number of egg masses was counted and females were dissected to determine their mating status. Adult survivorship on these trees was studied by placing virgin pairs of moths in two sleeve cages on each tree on 12 August. Adult mortality was recorded daily.

The effect of application rate on mating and oviposition was studied by treating four replicated four-tree plots in the Fuji orchard at the experimental farm with the three rates of M96–018 (0.06, 0.03, or 0.015 kg/l water). Trees were sprayed on 25 July, 8 and 22 August, and 5 September. Sleeve cages with one pair of virgin moths were placed on each tree and on 16 untreated trees on 1 and 15 August and 12 September. The mating status of females (presence or absence of a spermatophore) and the number of egg masses deposited were evaluated after 5 d in each test.

Choice oviposition tests were conducted with virgin moths released into screened cages (2 by 2 by 2.5 m) with pairs of potted 12-yr-old Red Delicious apple trees. One tree in each cage was sprayed with the standard rate of M96–018 (0.03 kg/l) and the other tree was sprayed with water. Trees were allowed to dry overnight, and 10 pairs of virgin moths were released into the cage the following morning. Tests were conducted on 8 and 22 July and 2 and 15 September. Two replicates were included on each date. Both trees in each cage were inspected for egg masses after 10 d.

Residual and Topical Effects of M96–018 on Eggs. Twenty potted apple trees were placed inside a screened building (6 by 20 by 4 m), and 60 pairs of virgin moths were released on 12 June. After 5 d, all trees were inspected for egg masses and the location of each egg mass was flagged. Ten trees with flagged egg masses were then sprayed with the standard rate of M96–018 (0.03 kg/l) on 18 June, and 60 additional pairs of moths were released on 19 June. After 5 d, all trees were again visually inspected for egg masses. Egg masses were collected and reared in the laboratory at 24°C. The number of larvae eclosing per egg mass was counted.

Residual Effects of M96–018 on Larval Development and Survivorship. Studies were replicated three times in a greenhouse to evaluate the development of larvae on apple seedlings treated either with M96–018 or left untreated. Five apple seedlings on each date were sprayed either with the standard rate of M96–018 (0.03 kg/liter) or water. Leaves from each treatment were collected weekly. Leaf petioles were placed in water-filled vials sealed with parafilm. Individual leaves were placed inside waxed 250-ml paper cups. Ten-d-old third-instars were placed individually on leaves and maintained at 24°C. Larvae were transferred to new leaves from each treatment every 7 d. The proportion of larvae pupating and the pupal weight for each sex was recorded.

The effect of spray application rate and residue concentration on larval survivorship and larval development was studied in a series ($n = 4$) of choice and no-choice bioassays. All leaves were collected from Fuji trees at the Moxee orchard sprayed on 8 and 22 August and 5 September with three rates of M96–018 (0.06, 0.03, or 0.015 kg/liter water) or from unsprayed trees. Leaves were collected on 8 and 15 August and 5 and 12 September for the no-choice assays. Similarly, leaves for the choice bioassays were collected on 5 and 28 August and 5 and 12 September. A single neonate larva was placed in a 50-mm plastic petri plate (Falcon, Lincoln Park, NJ) with two 10-mm leaf disks and kept at 24°C. Leaf disks in the choice bioassays included one untreated leaf disk and one disk from the particle film treatment. The larval feeding position and survivorship was recorded after 5 d in the choice bioassay. Larval weight and survivorship were recorded after 8 d in the no-choice bioassays. Sample size was 20 larvae in each test on each date.

Field Efficacy Trials. The efficacy of M96–018 to control the overwintering generation of *C. rosaceana* was investigated in three field trials with a randomized complete block design. The first spray trial was conducted in the Brewster Fuji orchard. Ten replicated 10-tree plots were treated with a single application of M96–018 with the backpack sprayer on 26 March. No green foliage was present in the orchard on this date. Shoots were visually inspected on 6 May for the presence of live leafrollers. In the second field test, we evaluated the effectiveness of M96–018 applied just before the beginning of bloom. A single application was made on 13 May in the Golden Delicious block at the experimental farm to 10 replicated single-tree plots. Populations of leafrollers on the treated and on 10 untreated trees were counted on 30 May. A third spray trial compared the efficacy of applying M96–018 just before bloom (8 May) versus at petal fall (16 May). This test was conducted in the Fuji block at the research farm. Five four-tree replicates were included for each spray timing plus an untreated control. The number of live larvae in each replicate was counted on 30 May.

Statistical Analyses. All count and proportion data were transformed square root ($x + 0.01$) and arcsine (square root [x]), respectively; before conducting the analysis of variance (PROC GLM, Hintze 1987). Means were separated with least significant difference where significant differences occurred. The Fisher exact test was used to compare the proportion of females mated in sleeve cages between M96–018-treated and untreated foliage and the position of larvae in the petri plate choice feeding tests.

Results

Residual Effects of M96–018 Against Adults. The proportion of females mated was significantly lower ($P < 0.05$, Fisher exact test) on the trees treated with the standard rate of M96–018 than on untreated trees for both July tests (Table 1). The mean number of egg masses also was significantly lower on the M96–018-treated trees compared with the untreated trees for both dates (3 July: $F = 36.9$; df = 1, 22; $P < 0.001$: 22 July: $F = 10.1$; df = 1, 13: $P < 0.01$). Significant differences in particle film density on leaves were detected in August and September among each of the three rates of M96–018 applied ($F = 62.1$; df = 2, 6; $P < 0.001$) (Table 2). Similar to the July tests, mated females laid fewer egg masses on trees sprayed with the three rates of M96–018 versus untreated trees ($F = 4.3$; df = 3, 8; $P < 0.05$). However, no difference in oviposition was found among the three rates (Table 2). Unlike the July tests, M96–018 did not affect the mating success of females compared with the untreated trees ($F = 2.2$; df = 3, 8; $P = 0.17$).

Table 1. Effects of repeated applications (0.03 kg/l) of particle film M96–018 on mating and oviposition of *C. rosaceana* in sleeve cages

Treatment	Date	Proportion of females mated	Mean no. egg masses laid per mated female (SE)
M96-018	3 July	0.28a	0.3 (0.2)a
Untreated	3 July	0.93b	1.7 (0.2)b
M96-018	22 July	0.25a	1.0 (0.3)a
Untreated	22 July	0.67b	2.4 (0.3)b

Proportion of females mated and mean number of egg masses per mated female on each date followed by a different letter are significantly different, $P < 0.05$.

Both sexes in sleeve cages with untreated shoots lived longer on average (0.33 and 0.55 d for males and females, respectively) than those placed on M96–018-treated shoots (sex: $F = 29.3$; df = 1, 75; $P < 0.001$; treatment: $F = 5.8$; df = 1, 75; $P = 0.02$). The interaction of sex and treatment was not significant ($P = 0.55$). The low longevity of these laboratory-reared moths (<4 d) observed in these tests was probably caused by the occurrence of maximum temperatures >30°C. In choice tests with potted trees in screened cages, *C. rosaceana* laid on average significantly more egg masses (mean ± SE) on untreated (4.0 ± 0.80) versus M96–018-treated (0.63 ± 0.26) trees ($F = 16.0$; df = 1, 14; $P = 0.001$).

Residual and Topical Effects of M96–018 on Eggs. The number of egg masses laid on potted trees after the application of M96–018 to one half of the trees in the screened building was more than three times higher on the untreated versus treated trees (Table 3).

Table 2. Effect of spray application rate of M96–018 on density of residues (mg/cm^2) on Fuji trees and on mating and oviposition of *C. rosaceana* in sleeve cages

Application rate, kg/l	Mean (SE) residue concn., mg/cm^2	Proportion (SE) of females mated	Mean no. egg masses laid per mated female (SE)
Untreated	—	0.79 (0.06)a	1.28 (0.08)a
0.015	0.10 (0.01)a	0.58 (0.10)a	0.69 (0.31)b
0.03	0.18 (0.01)b	0.44 (0.13)a	0.51 (0.12)b
0.06	0.29 (0.01)c	0.52 (0.13)a	0.48 (0.08)b

Column means followed by a different letter are significantly different, $P < 0.05$.

Table 3. Mean larval hatch from egg masses topically treated with M96–018 (0.03 kg/l) and from egg masses laid on treated residues on potted Delicious apple trees placed in a screened building

Treatment	No. egg masses	Mean no. larval eclosing per egg mass (SE)
Untreated	22	85.3 (18.70)a
Egg mass laid on M96–018	6	98.8 (36.74)a
Egg mass laid under M96–018	23	90.4 (21.78)a

Mean number of larvae for each treatment followed by the same letter are not significantly different, $P > 0.05$.

Table 5. Effects of the spray rate and leaf residue of M96–018 on larval weight and survivorship of *C. rosaceana* larvae reared on treated and untreated apple leaf disks for 10 d at 24°C

Application rate, kg/l	Mean (SE) residue concn., mg/cm^2	Mean (SE) larval wt, mg	% (SE) of larvae surviving
Untreated	—	3.9 (0.9)a	97.5 (2.5)a
0.015	0.13 (0.02)b	1.3 (0.4)b	61.5 (8.8)ab
0.03	0.25 (0.06)ab	0.8 (0.2)b	51.3 (10.1)b
0.06	0.40 (0.08)a	0.9 (0.3)b	46.8 (12.0)b

Column means followed by a different letter were significantly different, $P < 0.05$.

No significant effect on larval hatch was found for eggs laid beneath or over residues compared with untreated foliage ($F = 0.05$; df = 2, 48; $P = 0.95$) (Table 3).

Residual Effects of M96–018 on Larval Development and Survivorship. Significant effects were found in rearing third instars on apple seedlings treated with and without M96–018 in a greenhouse (Table 4). The percentage of larvae completing development and pupating was significantly lower on M96–018-treated plants ($F = 8.4$; df = 1, 4; $P < 0.05$). Lower pupal weights were found for both sexes developing on the M96–018-treated plants (male: $F = 29.2$; df = 1, 32; $P < 0.001$; female: $F = 65.3$; df = 1, 24; $P < 0.001$).

The effect of spray concentration on larval growth and survivorship was significant in no-choice tests (Table 5). The mean residue levels varied significantly among spray rates ($F = 5.5$; df = 2, 9; $P < 0.05$) and increased 1.6–1.8 times with each twofold increase in the spray rate. However, residue levels were not significantly different at the two lower spray rates in these tests. In the no-choice tests, a significant effect was found for larval survivorship ($F = 4.42$; df = 3, 12; $P < 0.05$). Significantly fewer larvae survived feeding on the leaf disks sprayed with the 0.03 or 0.06 kg/l rate. All three rates of M96–018 significantly reduced larval weight compared with larval feeding on untreated leaf disks ($F = 8.8$; df = 3, 12; $P < 0.01$) (Table 5).

In choice tests, the mean residue levels varied significantly among each of the three spray rates ($F = 23.5$; df = 2, 9; $P < 0.001$) and increased 1.7–1.8 times with each twofold increase in the spray rate. However, the spray rate did not significantly affect the feeding location of larvae in these tests ($F = 0.52$; df = 2, 9; $P = 0.61$), and no difference in larval survival among rates was found after 5 d ($F = 2.3$; df = 2, 9; $P = 0.15$). The data for larval feeding position were grouped across all three rates to compare larval choice of feeding on treated versus untreated leaf disks. Significantly fewer larvae were found feeding on M96–018-treated leaves than on untreated leaves ($P < 0.05$, Fisher exact test).

Table 4. Effects of treating apple seedlings in a greenhouse with 0.03 kg M96–018 per liter water on *C. rosaceana* larval survivorship and pupal weight

Treatment	% (SE) larvae pupating	Mean (SE) pupal wt, mg	
		♂♂	♀♀
Untreated	83.3 (3.3)b	52.3 (2.3)b	89.5 (3.9)b
M96–018	48.3 (11.7)a	33.0 (2.7)a	49.3 (2.3)a

Column means followed by a different letter were significantly different, $P < 0.05$.

Field Efficacy Trials. Apple trees sprayed with M96–018 at a delayed dormant timing in March had significantly fewer larvae (mean ± SE) feeding per tree in May than trees left untreated (0.4 ± 0.2 versus 1.6 ± 0.3 larvae per tree, respectively) ($F = 8.76$; df = 1, 18; $P < 0.01$). In contrast, larval density (mean ± SE) on trees sprayed with M96–018 at petal fall in the Golden Delicious orchard was not significantly different from larval densities on unsprayed trees (2.7 ± 0.3 versus 2.5 ± 0.3, respectively) ($F = 0.17$; df = 1, 18; $P = 0.68$). Similarly, there was no significant difference in larval population density (mean ± SE) on trees sprayed with M96–018 either before or after bloom (6.6 ± 0.8 and 4.4 ± 1.2, respectively) versus larvae on unsprayed trees (5.4 ± 0.4) in the Fuji orchard ($F = 1.75$; df = 2, 12; $P = 0.22$).

Discussion

Our results with the M96–018 particle film formulation against several life stages of *C. rosaceana* demonstrate that this new technology may be an effective management tool for this pest and perhaps other similar tortricid leafroller pests in tree fruit. In our tests, M96–018 had significant negative effects on adult survivorship, longevity, oviposition; and larval feeding and survivorship. We observed that oviposition on treated plants occurred only on leaves with low levels of residue. However, no effect was found on larval hatch when eggs were laid on M96–018 residues or when the product was applied topically to the egg masses. The mixed results we observed on the mating success of adults (Tables 1 and 2) were probably affected by our observations that some of the moths rested and mated on the untreated sleeve cage material in these tests. The significant reductions in oviposition that occurred on particle film-treated plants in all of the tests suggests that adult emigration from entire orchard blocks treated with particle films could be affected. However, the reduction in adult longevity for moths confined to particle film residues also may have contributed to the observed reduction in oviposition.

Field trials with M96–018 showed that particle films can be effective in controlling larval populations of *C. rosaceana.* A single spray applied before the initiation of budbreak in the spring reduced the number of larvae feeding inside leaf shelters by 75%. Larvae overwinter as second and third instars inside silken hibernacula under bud scales and in bark crevices and must disperse unknown distances to feed on vegetative and floral buds in the spring. The particle film adheres very strongly to bark and must in some way interfere with larval dispersal and settling in buds. However, the material had no effect on the density of larvae feeding inside rolled leaf shelters when it was applied before or after bloom because the foliage inside the shelter was not coated, yet the significant effects we observed on larval growth and survivorship in the greenhouse studies suggest that a more reliable measure of the impact of particle films in these field tests would have been if we had measured the number of pupae that developed in spray plots. *C. rosaceana* larvae generally feed on recently expanded leaves on shoot terminals that they roll with silk. Exposure of these larvae to residues inside these shelters is problematic. However, later instars of *C. rosaceana* build several shelters before pupating and thus are exposed to spray residues (Onstad 1985).

Management of leafrollers in tree fruits will require season-long coverage of new foliage, with multiple spray applications. The half-life of residues on leaves in our study was 7–14 d and we did not measure any increase in residue on foliage following repeated applications every 14 d (Unruh et al. 2000). Continual erosion of residues occurred because of abrasion among plant parts, wind, and precipitation. Season-long use of M96–018 with 7–10 applications of the standard rate applied every 14 d resulted in a 63% reduction in fruit damage by the fruittree leafroller, *Archips argyrospilus* (Walker), in a replicated small-plot study in apple in 1998 (A.L.K., unpublished data). Similar field trials are needed to evaluate the effectiveness of particle films for *C. rosaceana.*

The range of effects of M96–018 we observed with *C. rosaceana* are similar to the results reported by Glenn et al. (1999) for several important leaf feeding apple and pear pests. For example, they found that pear psylla adults in choice tests did not settle on or oviposit on particle film-treated surfaces. Similarly, potato leafhopper populations did not develop in a greenhouse on treated apple seedlings, whereas populations on untreated seedlings caused severe plant damage. Mortality of pear psylla adults confined to leaf-treated surfaces was higher than those on untreated leaves, and populations of the spirea aphid and spider mites experienced 50% higher mortality than populations on untreated plants.

Despite these significant effects on a range of pest species, it is not clear how particle films should be used in apple and pear pest management. Clearly, a water soluble formulation that can be applied with conventional spraying equipment would be a big improvement in handling the material. The effects of particle films on fruit quality need to be evaluated, and the removal of residues from fruit after harvest is an important concern with this product. The material appears to be ideally suited for pests that are feeding on mature leaves (psylla and leafhoppers). Pest species that feed on newly expanding foliage or are protected from exposure to residues may be more difficult to control (aphids, codling moth, leafrollers, mites, leafminers).

Dust particles are well-known to be disruptive of natural enemies (DeBach 1979) and resurgence of pest species populations that are regulated by parasitoids (scale, leafminers) or predators (phytophagous mites) may occur in particle film-treated orchards. For example, the population density of the western tentiform leafminer, *Phyllonorycter elmaella* Doganlar & Mutuura, in the Fuji orchard at Moxee were two to five times higher in the plots treated with the three rates of particle film than on the untreated trees on 8 September (A.L.K., unpublished data). Larval parasitism levels in these particle film plots averaged <20% compared with 74% on the untreated trees. Interestingly, leafminer density in these trials was inversely related to the rate of spray application of the particle film. These data suggest that the particle film adversely affected the parasitism of *P. elmaella,* and that the higher densities of particle film may impact leafminer oviposition or egg survivorship. Further evaluation of M96–018 and other particle film formulations will require assessment of the impacts of various single or multiple-spray programs on the large and varied suite of tree fruit pests and their natural enemies.

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