Moisture tempers impairment of adult *Otiorhynchus sulcatus* (Coleoptera: Curculionidae) climbing ability by fluoropolymer, talc dust, and lithium grease

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ABSTRACT

As part of a project to develop tools for the physical exclusion of flightless root weevils, adult black vine weevils (BVW), *Otiorhynchus sulcatus* (F.), were placed in open enclosures with smooth walls of glass, plastic or aluminum to test their ability to escape by climbing. Enclosure walls were left untreated or were treated with substances known to reduce insect climbing ability: fluoropolymer, powdered talc and lithium grease. No BVW escapes were observed under dry conditions, but all treatments allowed some escapes under wet conditions, suggesting that moisture helps BVW adults scale treated surfaces. The results help explain the ability of root weevils to overcome physical barriers under field conditions.

Key Words: black vine weevil, insect barrier, physical control, root weevil

INTRODUCTION

Like other root weevils, the black vine weevil (BVW), *Otiorhynchus sulcatus* (F.), feeds on roots as a larva, leaves as an adult and disperses by walking during the wingless adult phase. The biology and control of BVW was reviewed by Moorhouse *et al.* (1992).

Flightless root weevils could be particularly susceptible to physical control by exclusion. While hardly a new strategy (Feytaud 1918), physical control has recently been the subject of some interest (Vincent *et al.* 2003). An aluminum fence with a band of lithium grease (Cowles 1995, 1997) or fluoropolymer-coated tape (Bomford and Vernon 2005) near the upper edge can limit root weevil movement. Also effective is a portable plastic trench, designed to exclude Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Hunt and Vernon 2001). Both the fence and the trench have reduced root weevil immigration into strawberry plots by about two-thirds (Bomford and Vernon 2005). Sticky bands and fluoropolymer-coated tape on shrub stems are both recommended to reduce adult feeding on leaves (Antonelli and Campbell 2001).

Like other insects, root weevils climb using a combination of tarsal claws to hook textured surfaces and adhesive pads on their tarsomeres to attach to smooth surfaces. These adhesive pads consist of densely packed setae, each with a terminus a few µm in diameter that attaches to the surface through weak van der Waals and capillary forces (Arzt *et al.* 2003, Gao and Yao 2004). The sum of these weak forces can support the insect only if a sufficient proportion of the setae contact the surface.

Insect tarsi cannot adhere to surfaces with sufficient micro texture to prevent a large proportion of setae from making contact, but insufficient macro texture for tarsal claws to grip. Lithium grease is one such surface, consisting of an open, fibrous crystal matrix that holds tiny (~1 µm) oil droplets (Wilson 1964); fluoropolymers have similar properties (Hougham 1999). Smooth surfaces coated with fine, loose

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dust particles are similarly difficult for insects to climb because their tarsi adhere to dust particles, which slip away from the surface (Boiteau and Vernon 2001). Smooth dusted surfaces have shown potential as physical barriers to Colorado potato beetle (Boiteau *et al.* 1994, Boiteau and Osborn 1999), and root weevil (M.K.B., personal observation) movement.

This paper describes laboratory and field studies testing the influence of surface treatment and moisture on the ability of adult BVW to climb materials that could be used to construct physical barriers to root weevil migration. The results are intended to aid in the development of physical control tactics for root weevil management.

MATERIALS AND METHODS

Test insects. BVW adults were collected from an apple rootstock nursery and home garden near Vancouver, BC in late summer and early fall. Weevils were held for no more than 30 days at 20 ºC under a 16:8 h L:D photoregime in clear plastic cages containing potted strawberry, *Fragaria x ananassa* (Duchesne) plants as a food source.

Glass surface treatments (dry). Eleven 250 mL glass Erlenmeyer flasks were washed and dried. One end of a length of surgical tubing was placed in each flask to allow air to escape as it was dipped upside-down in liquid fluoropolymer (Insect-A-Slip, BioQuip, Rancho Dominguez, CA) (four flasks), or powdered talc (four flasks), evenly coating the top 3 cm of the neck with the dip treatment. Excess talc and fluoropolymer were shaken off, and the fluoropolymer was allowed to dry to a hard, smooth finish. Three remaining flasks were left untreated as controls (unequal replication reflects flask availability). Flasks were randomized, five BVW adults were placed in each and all flasks were placed in an incubator held at 20 ºC and 20% RH under a 16:8 h L:D photoregime. The number of weevils in each flask was recorded after 0.5 h and all escapees were removed from the incubator. The number of weevils remaining in each flask was recorded again after 24 h when the experiment was terminated. Data were analyzed by one-way ANOVA for unequal number of replicates and treatment means were separated by Tukey's honestly significant difference test (JMP Version 4.0.4, SAS Institute 2001).

Outdoor plots. Three, one m square enclosures, constructed from aluminum gutters (75 mm deep by 120 mm wide) sealed at all joints with hot glue, were sunk into freshly-tilled soil so that the soil surface was even with the upper lip of the gutter. The soil inside each enclosure was covered with a square of landscape fabric with its edges screwed to the inner gutter wall. One litre of 1:1 water:dormant oil emulsion was poured into each gutter.

Each enclosure was randomly assigned to one of three treatments: The landscape fabric pad was separated from the gutter by: 1) a 20 cm high aluminum fence with fluoropolymer-coated tape (EnviroSafe, Professional Ecological Services, Victoria, BC) attached to the upper edge of the inner surface (fence); 2) a portable plastic trench (Hunt and Vernon 2001) coated inside with dormant oil (trench); or 3) no barrier (control).

Two days after plot setup, marked BVW adults were released in the centre of each enclosure at 2200 h, a time of high activity among wild specimens observed in the area. A flashlight was used to observe weevil movement at five min intervals for one h after release. Weevils that entered the aluminum gutter and became trapped in the dormant oil emulsion (successful escapes) were recorded during the first hour and again the following morning at 1000 h. The experiment was conducted in the same plots three times (13, 18, and 20 August 1997), with ten BVW per treatment in the first replicate and 20 in the others. Hourly RH readings recorded at the Vancouver International Airport (6 km from study site) during each observation period were used to estimate the ambient RH range for each replicate (Environment Canada 2005).

A two-way ANOVA was used to test for treatment and replicate effects on weevil escape rates after one and 12 h, and for interaction between factors (JMP Version 4.0.4, SAS Institute 2001). Means were separated by Tukey's HSD test.

Plastic surface treatments (wet vs. dry). Forty, 35 mL black plastic film canisters (30 mm diameter by 50 mm deep) were washed, dried, and randomly assigned to one of four treatments: ten were untreated controls; ten were dusted with powdered talc; ten were coated with liquid fluoropolymer; and ten had a 2.5 cm band of white lithium grease applied to the inner top edge.

The following day, half of the canisters from each group were rinsed with water and then emptied, leaving droplets inside. These were placed in a sealed plastic container containing an open water source to create a saturated environment. The remaining unrinsed canisters were placed in an identical container without a water source (ambient RH: 50-74%, Environment Canada 2005) and left open to allow air circulation. Canister order was randomized within each container.

Two BVW adults were placed at the bottom of each canister. The number of weevils remaining in each canister was recorded and escapees were removed at 0.5 h intervals for 3.5 h. Canisters were not treated on the outside, so re-entry was possible, but never observed. ANOVA was used to test for treatment effects within each container and means were separated by Tukey's HSD test (JMP, Version 4.0.4, SAS Institute 2001). A t-test was used to compare escape rates between containers for each treatment.

Plastic surface treatments (saturated vs. ambient RH). Eighteen, 290 mL plastic cups (50 mm diameter at base, 70 mm diameter at opening, 100 mm deep) were randomly assigned to one of three treatments: six were untreated controls; six had a 2.5 cm strip of white lithium grease applied around the inner top edge; and six were dusted with powdered talc.

Three BVW adults and a moist cotton swab were placed in the bottom of each cup. Cups from each treatment were evenly divided into two identical plastic tubs, each containing a damp cloth. One tub was sealed to create a saturated environment in which condensation formed on the plastic cups; the other tub was left open to allow air circulation and prevent condensation (regional ambient RH: 67-95%, Environment Canada 2005). Tubs were held at 20 °C for 20 h. Any weevils that escaped from their cups were removed from the tubs at hourly intervals for the first six hours and then every other hour thereafter until the study was terminated. The mean number of escapes per cup was calculated for each treatment in the open and sealed containers. ANOVA was used to test for treatment effects within each container and means were separated by Tukey's HSD test (JMP Version 4.0.4, SAS Institute 2001). A t-test was used to compare escape rates between containers for each treatment. The time required to escape under each combination of conditions was estimated by Kaplan-Meier analysis and a Wilcoxon t-test was used to test for differences in escape times between treatments (JMP Version 4.0.4, SAS Institute 2001).

RESULTS

Glass surface treatments (dry). Almost all (93.3 ± 3.3%, $n = 3$) weevils in the control flasks escaped, but none (0.0 ± 2.9%, $n = 4$) escaped from flasks treated with talc dust or fluoropolymer, demonstrating a strong treatment effect ($F_{2,8} = 285$, $P < 0.001$). All escapes from the control flasks occurred within the first 30 min of the 24 h observation period. Weevils in the fluoropolymer treated flasks were frequently observed walking up the glass to the fluoropolymer strip and were occasionally able to climb part-way over this strip before falling. When the experiment was terminated, approximately half of the weevils in the fluoropolymer treated flasks were on the flask walls. Weevils in the talc treated flasks showed much less ability to scale the glass walls and were all at the bottom of the flask at the end of the experiment.

Outdoor plots. Treatment and replication both affected weevil escape rates ($F_{2,141}$ = 189 and 25, respectively; $P < 0.001$) and an interaction was found between these factors ($F_{4,141} = 12$; $P < 0.001$). Almost all weevils left control plots over the course of all replications (Figure 1), but escapes from plots surrounded by physical barriers only occurred in the third replication, conducted under light rain and high humidity conditions. Under the drier conditions of the first two replications weevils quickly climbed the aluminum fence to the lower edge of the fluoropolymer-coated tape and were unable to climb further for the duration of the test. Most weevils surrounded by plastic trenches fell into the trenches and none emerged. Under the wet conditions of the third replication the first of 20 weevils was able to walk onto the fluoropolymer within 5 min of its release. Within 20 min, four more had achieved this feat, two had reached the top of the aluminum fence and one had crossed the trench. Statistical comparison of the replications showed a higher escape rate from the fenced treatment in the third repetition after 12 h ($F_{2,47} = 26$; $P < 0.001$), but not from the trenched treatment ($F_{2,47} = 2.5$; $P = 0.09$).

Plastic surface treatments (wet vs. dry). Under dry conditions all weevils escaped from untreated canisters but none escaped from those treated with talc, fluoropolymer, or white lithium grease (Table 1). Talc lost its dusty character under wet conditions, allowing more escapes (Table 1). The dried fluoropolymer reverted to a liquid state in the presence of moisture, clumping on tarsi and allowing only one

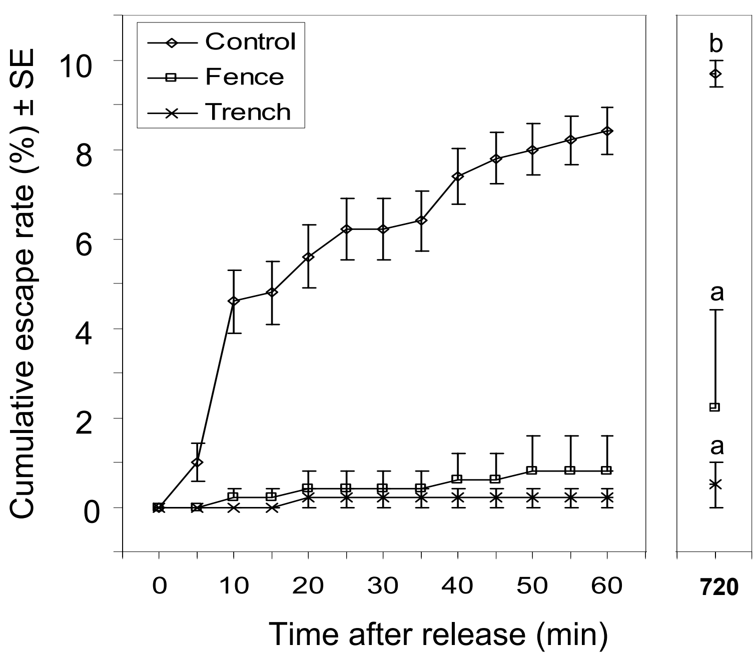

Figure 1. Black vine weevil escapes from a one m square area surrounded by a 20 cm high aluminum fence with fluoropolymer-coated tape attached inside (fence), a portable exclusion trench (trench) or no barrier (control). Observations were made at 5 min intervals for 1 h after insect release and 12 h after release. Lower error bars omitted from fence data points for clarity. Final means labeled with the same letter do not differ significantly at $\alpha = 0.05$ (Tukey's HSD test, $n = 3$).

Table 1.
Mean percentage of adult black vine weevils that left plastic canisters or cups that were untreated (control) or coated inside with dried fluoropolymer, white lithium grease (grease) or powdered talc (talc). Canisters and cups were placed in an open container (ambient RH) or a closed container with an open water source (saturated RH). Canisters were rinsed immediately before being placed in the closed container, leaving their surface wet.

	Canister escapes (%)[1], $n = 10$			Cup escapes (%)[1], $n = 9$		
Treatment	Dry surface, ambient RH	Wet surface, saturated air		Dry surface, ambient RH	Dry surface, saturated air	
Control	100 A	90 a A	$t_{18} = 2.3, P = 0.15$	89 a A	100 a A	$t_{16} = 1.0, P = 0.33$
Fluoro-polymer	0 A	10 b A	$t_{18} = 2.3, P = 0.15$	-	-	
Grease	0	0 b		0 b A	33 b A	$t_{16} = 4.0, P = 0.06$
Talc	0 B	70 a A	$t_{18} = 73, P < 0.0001$	0 b	0 c	
		$F_{3,39} = 50$		$F_{2,24} = 64$	$F_{2,24} = 38$	
		$P < 0.0001$		$P < 0.0001$	$P < 0.0001$	

[1] Means followed by the same lower case letter within a column do not differ significantly (Tukey's test, a= 0.05); those followed by the same upper case letter within a study and row do not differ significantly (t-test, a= 0.05).

escape. Weevils in fluoropolymer-treated canisters largely ceased their activity until the experiment ended. No weevils escaped from moistened grease-treated canisters.

Plastic surface treatments (saturated vs. ambient RH). Visible condensation first appeared on cups in the saturated environment 8 h after the test began and was very heavy by the end of the test. No condensation was seen on cups in the lower humidity environment. Almost all weevils escaped from control cups within the first hour of observation; the only weevil that did not escape from a control cup in an hour did not escape at all (Table 1). No weevils escaped from cups treated with talc in either container. One third of the weevils escaped from grease-treated cups in the saturated environment, but none escaped in the ambient RH environment (Table 1). On average, escapes from greased cups took longer than escapes from untreated cups in the sealed container (16.7 ± 0.7 versus 0.7 ± 0.7 h, respectively; $\chi^2 = 27$, df = 2; $P < 0.001$). Mean escape times from untreated cups did not differ between the open and sealed containers.

DISCUSSION

Under dry conditions talc dust, fluoropolymers and lithium grease treatments rendered several smooth surfaces (glass, plastic, and aluminum) unclimbable to BVW adults for the duration of our tests. Equivalent treatments were sometimes less effective under wet conditions, or in saturated environments. This may help explain why physical barriers that would be expected to offer total exclusion, based on observations under dry conditions, exclude only two-thirds of root weevils in the field (Bomford and Vernon 2005).

Most adult weevils quickly attempted to leave the open containers we used for our tests. Their success in exiting, and the length of time they took to leave, were considered indicators of the difficulty they had in scaling the barriers they faced. Under dry conditions, surface treatments eliminated escapes; under wet conditions they usually reduced the proportion of insects able to escape and lengthened escape times.

Cowles (1995) has suggested that root

weevils are able to evade physical barriers because natural bridges form over otherwise unclimbable surfaces. He has seen field debris, such as twigs, adhering to the white lithium grease on his barriers, and plant canopies touching across barriers (R.S. Cowles, pers. comm., see Acknowledgements). We have also seen natural bridges that could allow root weevils to cross portable trench barriers in field studies (Bomford and Vernon 2005), but these were not a factor in the tests reported here.

We observed repeated instances of BVW adults crossing vertical surfaces treated with fluoropolymer, talc dust, and lithium grease in the presence of moisture. BVW adults scaled talc-dusted plastic that had been lightly rinsed to mimic rainfall on a dusted plastic exclusion trench. Similar observations have been reported previously for Colorado potato beetles challenged by plastic-lined trenches after rainfall in field studies (Boiteau *et al.* 1994). Rinsing did not render greased surfaces climbable in one test, reflecting field observations in which greased aluminum barriers excluded root weevils after irrigation (Cowles 1995). We did, however, observe BVW scaling greased plastic with visible surface condensation in a high humidity environment and scaling fluoropolymer-treated aluminum in a light rain shower. We are unaware of other reports of moisture enhancing an insect's ability to scale fluoropolymer or lithium grease-coated surfaces. These observations lead us to suggest that the insects' tarsal pads adhere to condensation on treated surfaces. Essentially we hypothesize that the insects can overcome physical barriers by walking on water.

More rigorous tests of this hypothesis are necessary. The studies reported here reflect a variety of treatment combinations observed under different conditions. Experimental factors were sometimes confounded. For example, BVW were unable to scale a fluoropolymer treated fence under dry conditions two and seven days after the fence was erected, but scaled the same fence in a light rain shower nine days after setup. We attributed this difference to the presence of moisture, but it might also have been an effect of fence age. Similarly, our analyses of interactions between surface treatment and environment were confounded by the fact that surface treatments were replicated within environments, but only one instance of each environment was tested in any study. Our observations suggest intriguing avenues for further study, not definitive conclusions.

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