# Plant Essential Oils as Arrestants and Repellents for Neonate Larvae of the Codling Moth (Lepidoptera: Tortricidae)

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**ABSTRACT** Nonhost chemicals may be useful for controlling insect pests of crop plants by interfering with orientation to, and selection of, host plants. Essential oils of 27 plant species were tested in 2 different laboratory assays for evidence of arrest and repellency of neonate larvae of the codling moth, *Cydia pomonella* L. In an olfactometer in which larval upwind movement toward apples was assessed, greatest arrest was achieved with oils of lavender, *Lavandula officinalis* L.; pennyroyal, *Mentha pulegium* L.; and cypress, *Cupressus sempervirens* L.. Oil of lavender was most effective in preventing larvae from moving upwind in the olfactometer. In a barrier assay, essential plant oils were applied to the distal ends of a glass rod (15 cm long) on which larvae were placed. Larvae crossed the barrier to reach apples impaled on each end of the glass rod. The most effective repellents in this barrier assay were rue, *Ruta graveolens* L.; garlic, *Allium sativum* L.; patchouly, *Pogostemom cablin* (Blanco); and tansy, *Tanacetum vulgare* L., oils. These 4 plant essential oils were ingredients, may be useful in protecting fruit from attack by codling moth larvae by preventing larvae from orienting to and arriving at fruit.

KEY WORDS Cydia pomonella, repellent, deterrent, arrestant, attractant

THE CODLING MOTH, Cydia pomonella L., is a cosmopolitan pest of apples and other fruits. In many areas it is the principal cause of damage to fruit of apple; Malus X domesticus Borkh; and pear, Pyrus communis L.) (Beers et al. 1993), and control of this insect is key to any integrated pest management (IPM) system. New methods are needed to control this pest and reduce our reliance on broad-spectrum pesticides because of increasing resistance in codling moth to certain pesticides (Varela et al. 1993, Knight et al. 1994) and increasing restrictions on pesticide use on agricultural crops. Mating disruption using the codling moth female sex pheromone is successful in some areas but requires relatively low moth population densities for success (Carde and Minks 1995). Supplemental and compatible approaches would be helpful where population densities are too high for mating disruption to work.

Plants, including cultivated crop plants, may be protected from insect attack by the presence of a diversity of plants or by the presence of particular nonhost plants (Hill 1976; Latheef and Irwin 1979, 1980). For example it has been suggested that mixed garden plantings reduce or prevent insect damage (Rodale 1966, 1974). These effects may be caused by chemical masking or repellency of insects by nonhost plant chemicals (Atsatt and O'Dowd 1976). A plant-seeking insect may be less able to discern and respond to the odor of its host in the presence of additional plant odorants (masking) (Bernays and Chapman 1994). Herbivorous insects also may be repelled or arrested by chemicals from nonhost plants or by host chemicals at high concentrations. See Dethier (1960) for a discussion of the use of the terms arrestant and repellent.

The application of plant extracts or essential oils to a crop or commodity may provide some degree of protection against insect pests (Rodale 1966; Jilani and Su 1983; Tingle and Mitchell 1984, 1986); Klepzig and Schlyter 1999. These applications may be effective by an insecticidal mode, by arresting or repelling attracted insects, by host odor masking, or deterrence of feeding. Many plant extracts or chemicals have been evaluated as feeding deterrents and as insecticides by applying them to plant foliage or by incorporating them into artificial diets for various plant-feeding insects (Jacobson 1990). Comparatively little work has been done to evaluate nonhost chemicals as behavioral repellents or arrestants to prevent insect contact with the host plant.

The codling moth adult lays most eggs on host foliage, and larvae generally must find fruit on which to feed (Jackson 1978). Neonate codling moth larvae are attracted to the odors of apple fruit (Sutherland, 1972, Landolt et al. 1998). This behavior provides an opportunity to prevent newly hatched larvae from reaching and infesting fruit. We hypothesize that larvae may be prevented from reaching fruit by extracts of particular nonhost plants that are suitably effective as arrestants or repellents. We report here the comparative assessment of essential oils of 27 plant species as arrestants and repellents against neonate codling moth larvae. Using the definition of Dethier (1960) that an arrestant stops or slows movement, we considered a significant reduction in forward movement toward apple odor as evidence of arrest. Using the definition of repellent as causing orientation away from the source (Dethier 1960), we considered a significant rate of turning away from a plant oil as evidence of repellency. Either or both of these terms may be interpreted by others as deterrence. This work provides several candidate plant oils to develop in field trials as applications to reduce codling moth infestation of apple and other crops.

## Materials and Methods

**General.** Neonate codling moth larvae were used in all experiments. Eggs on wax paper were obtained from a laboratory insectary with an established codling moth colony. Three hours before the start of bioassays, larvae were shaken off wax paper strips with eggs and the wax paper strips were then placed in a plastic box. Larvae that subsequently emerged were 0–3 h old at the start of an experiment. Eggs and larvae were kept in a controlled environment room at 20°C, 50% RH and in a photoperiod of 14:10 (L:D) h.

Thinning apples used in bioassays were collected from apple trees (Red Delicious) in June of 1997 and were stored at 4°C and in darkness until needed. They were removed from storage and were kept in a laboratory at 20°C for 24 h preceding experiments. Apples used were uninfested, were not damaged, rotten or shriveled, and were 2-2.5 cm in diameter. Plant essential oils were purchased from Herbal Advantage, (Rogersville, MO), Pete's Naturals (Reading, PA), and Aromatherapy (Denville, NJ). Samples were initially quantified by weight and were then diluted in methvlene chloride to provide desired concentrations. Assays were conducted in a controlled environment room under fluorescent lighting, at 25°C and 40-60% RH. Experiments were conducted between 0900 and 1600 hours. Lights came on at 0600 hours and went off at 2000 hours.

Five experiments were conducted, 3 using an olfactometer design to test for arrest of larvae by odorants in an airstream and 2 using a barrier test with materials applied to a surface to test for repellency of larvae. With each assay design, 27 essential oils were evaluated at 1 dose to select materials for further testing. Additional experiments were then conducted, using both assay designs. These provided comparisons of different amounts of the most promising of those essential oils. One experiment also was conducted to assess how long the arrestant effects of an application might last in the olfactometer.

Olfactometer Tests. The olfactometer consisted of 2 parallel glass tubes through which air was forced. For each olfactometer tube, air was supplied from a small pump. Air was forced 1st through a charcoal filter, then through a flow meter at 180 ml/min, through a 500-ml glass jar that held the treatment, and finally into the olfactometer tube itself. The jars holding the treatments were fitted with a Teflon seal and steel bulkhead fittings for introducing and venting airflow. The glass olfactometer tube (1.4 cm inside diameter, 15 cm long) was fitted with a 17-gauge galvanized steel wire suspended within the length of it. Larvae were placed on the wire, at the downwind end of the tube, and were observed for 2 min. For each larva, the maximum distance upwind attained during the bioassay time was recorded.

To evaluate a plant oil, 3 thinning apples were placed in each jar of the olfactometer and a plant oil treatment was added to 1 jar and a solvent treatment (control) was added to the other jar. For the plant oil treatment, an application of 10 mg of oil in 100 ml of methylene chloride was made to filter paper (5.5 cm diameter, Whatman #3, Whatman, Hillsboro, OR) which was placed in the jar. For the solvent treatment (control), 100 ml of methylene chloride was applied to a filter paper that was placed in the other jar. Filter papers were held for 15 s before placement in a jar to allow the solvent to evaporate. After an initial wait of 2 min, larvae were placed in each of the 2 tubes and were monitored simultaneously for the 2-min assay period. Larvae were then removed and replaced with 2 more larvae (1 per tube), to be watched for 2 min. After 5 larvae were tested in each of the 2 tubes, the 2 holding jars, olfactometer tubes, and associated plumbing were switched. Five more larvae were then tested in each tube. All glassware and tubing downwind of, and including, the holding jars were then washed (hot soapy water, water rinse, acetone rinse, hexane rinse) and baked in an oven at 140°C for 2 h before use in additional assays. This test was replicated 5 times for each plant oil, providing 50 larvae tested per plant oil with thinning apples, with a corresponding 50 larvae tested to thinning apples alone.

Two plant oils that were effective in arresting codling moth larval upwind movement at 10 ml were then evaluated at amounts of 0, 0.1, 1, 10, and 100 mg. All dosages were applied in 100  $\mu$ l of methylene chloride to a filter paper 5.5 cm in diameter. The paper was then held in a fume hood for 30 s to allow the solvent to evaporate. For the assay, 1 olfactometer system contained 3 thinning apples, whereas the other system contained 3 thinning apples and a plant oil treatment. As before, 5 larvae were tested sequentially in each tube, the systems were switched, and 5 more larvae were tested sequentially per tube. This comparison was conducted 1st with the control or 0 dose, followed by the 0.1-mg dose, continuing with higher doses to complete the set. The complete set of assays was replicated 4 times for lavender oil, Lavendula angustifolia L., providing 40 larvae tested per treatment, and 5 times for pennyroyal oil, Mentha pulegium L., providing 50 larvae tested per treatment.

The most effective oil, determined in the 1st test described above, also was evaluated in an aging experiment to determine how the effectiveness of an application of oil to a filter paper in the olfactometer changed with time. A 10-mg dose was applied to a filter paper which was placed in a glass jar with 3 thinning apples. The other jar of the olfactometer contained 3 thinning apples and no plant oil. Five larvae were then tested for upwind movement in each tube of the ol-

	Plant name	Mean $\pm$ SE forward distance, cm		Paired $t$ -test <sup>a</sup>	
Common	Scientific	Treatment	Control	t	Р
Lavender	Lavendula angustifolia L.	$0.24\pm0.16$	$6.87\pm0.89$	7.49	< 0.001
Pennyroyal	Mentha pulegium L.	$1.07 \pm 0.46$	$10.68\pm0.80$	10.46	< 0.001
Cypress	Cupressus sempervirens L.	$1.10 \pm 0.44$	$3.43 \pm 0.70$	2.85	0.008
Lemon	Citrus limon Osbeck	$1.47 \pm 0.39$	$3.89 \pm 0.71$	2.90	0.007
Dill	Peucedanum graveolens Benth and Hook	$1.60 \pm 0.46$	$3.93 \pm 0.88$	2.45	0.020
Garlic	Allium sativum L.	$1.83 \pm 0.43$	$5.05 \pm 0.76$	3.42	0.002
Spearmint	Mentha spicata L.	$1.87 \pm 0.61$	$4.57 \pm 0.84$	2.59	0.015
Lavendin-S	Lavendula X intermedia Emeric ex Lois	$1.90 \pm 0.58$	$6.18 \pm 0.83$	3.70	0.009
Coriander	Coriandrum sativum L.	$2.08 \pm 0.52$	$4.35\pm0.78$	2.19	0.040
Balsam fir	Abies balsamea (L.) Miller	$2.13 \pm 0.73$	$3.67 \pm 0.83$	1.30	0.190
Wintergreen	Gautheria procumbens L.	$2.20\pm0.72$	$5.28 \pm 0.94$	2.75	0.010
Spruce	Picea mariana (Miller)	$2.47 \pm 0.76$	$5.37 \pm 0.88$	2.85	0.008
Basil	Ocimum basilcum L.	$2.52 \pm 0.59$	$4.23\pm0.87$	1.66	0.100
Juniper	Juniperus communis L.	$2.53 \pm 0.69$	$3.15 \pm 0.83$	0.50	0.600
Peppermint	Mentha piperita L.	$2.27 \pm 0.71$	$7.18 \pm 0.98$	4.70	< 0.001
Rosemary	Rosmarinus officinalis L.	$3.51 \pm 0.70$	$5.72 \pm 0.96$	2.04	0.050
Citronella	Cymbopogon nardus (L.)	$4.42 \pm 0.94$	$4.92 \pm 0.82$	0.40	0.690
Patchouly	Pogostemon cablin (Blanco)	$4.80 \pm 0.93$	$6.50 \pm 0.94$	1.67	0.100
Bitter orange	Citrus aurantium L.	$4.90 \pm 0.95$	$5.55\pm0.99$	0.51	0.620
Grapefruit	Citrus paradisi (MacFayden)	$5.42 \pm 0.92$	$5.30 \pm 0.86$	0.09	0.920
Ginger	Zingiber officinale Roscoe	$5.68 \pm 0.98$	$8.20\pm0.92$	2.08	0.040
Tansy	Tanacetum vulgare L.	$5.88 \pm 0.92$	$5.13 \pm 0.94$	0.75	0.460
Scotch pine	Pinus sylvestris L.	$7.01\pm0.86$	$5.93 \pm 1.04$	0.92	0.360
Sage	Salvia officinale L.	$3.10 \pm 0.80$	$6.58 \pm 1.03$	2.56	0.016
Eucalyptus	Eucalyptus globulus (Labille)	$4.32\pm0.87$	$5.88 \pm 0.91$	1.23	0.220
Tagetes	Tagetes glandulifera Schrank	$4.58 \pm 0.79$	$5.83 \pm 0.90$	1.10	0.280
Rue	Ruta graveolens L.	$4.07 \pm 0.88$	$3.78 \pm 0.82$	0.26	0.800

Table 1. Mean maximum forward distance attained by neonate codling moth larvae in parallel tube olfactometer in the 2-min bioassay period, in response to 3 thinning apples in the chamber (control), or 3 thinning apples plus 10 mg of plant essential oil on a filter paper (treatment)

<sup>*a*</sup> Results from paired *t*-test comparing responses for treatment versus control, df = 29.

factometer, using assay procedures described above. This was conducted 1, 3, 7, and 24 h after application. The entire sequence was repeated 6 times on 6 different days.

For each plant oil tested in the olfactometer, data for maximum distance upwind attained in response to the treatment and in response to the control were compared using a paired *t*-test. Data sets from comparisons of doses were subjected to linear regression analyses to determine if there were significant effects of dose on larval response. For each dose tested, data between treatment and control (attraction to 3 apples versus attraction to 3 apples plus plant oil) were compared, using a paired *t*-test. Response data for the aging experiment were also subjected to an analysis of variance (ANOVA) with means separated using the Tukey test.

**Barrier Assay.** A glass rod (3 mm diameter, 15 cm long) was imbedded at each end into thinning apples. A dilute solution of plant oil in methylene chloride (10 mg/ml) was applied with a small (#1) camel's-hair brush to the glass rod near each apple, providing a swath of oil  $\approx$ 4 mm wide. The application encircled the rod  $\approx$ 2 cm from the apple. Five neonate codling moth larvae were placed at the middle of the glass rod, and were observed until they all reached an apple or until 1 h elapsed. The time it took each larva to cross an application of oil and reach an apple was recorded, as was the number of times the 5 larvae turned around at the barriers. This assay was conducted 3 times with

each of the 27 plant oils screened. Between assays, glass rods were washed 3 times with methylene chloride and 3 times with acetone. Control tests, evaluating larval performance following applications of solvent to the glass rod, were conducted in the same manner daily.

The 4 plant oils most effective in repelling codling moth larvae at 10 mg of oil per milliliter of solution used in the barrier assay were selected for an assessment of a range of concentrations. The most effective repellents were indicated as those with the highest mean time required for larvae to cross the application made to the glass rod. Each plant oil selected was tested at 5 concentrations (0, 0.1, 1, 10, and 100 mg of oil per milliliter of methylene chloride). Data collected were times elapsed until a larva crossed a barrier and reached an apple (up to 1 h), and the number of times larvae turned around at a barrier within that time period, as evidence of repellency. Larvae were tested 5 at a time, with 3 replicates of this test conducted per concentration of each oil. Concentrations were tested in series, beginning with the lowest and ending with the highest concentration to minimize contamination risks. Glassware was cleaned as described above and apples were replaced between sets.

For each plant oil, both response time (time elapsed until the larva crossed a barrier) and number of turnarounds were compared for treatment and corresponding control data by a paired *t*-test. Data from comparisons of different concentrations were subjected to regression analyses to determine if there was a significant effect of concentration on either response time or numbers of turn arounds.

# Results

Olfactometer Tests. The 27 essential plant oils tested in the olfactometer varied in their effectiveness in arresting neonate codling moth larvae (Table 1). Arrest was indicated by a significant reduction in upwind movement in comparison with the untreated control (Table 1). Such effects were observed with 15 of the 27 plant oils tested. Lavender was noteworthy in keeping larval upwind movement to a minimum despite good responses by larvae to thinning apples in the control. A continuum of effectiveness was evident for the remaining 14 plant oils that showed some indication of arrest ( $P \le 0.05$ ; Table 1). For several plant oils, there was no indication of any effect (e.g., rue, Ruta graveolens L.; scotch pine, Pinus sylvestris L.; and tansy, Tanacetum vulagare L.). None of the plant oils showed a significant positive effect (attraction) on larvae. Pennyroyal and lavender were selected for further testing, using a comparison of doses of the oils.

The arrest of larvae in the olfactometer was correlated to plant oil dose with both lavender and pennyroyal oils, with the maximum effect attained with the highest dose tested (100 mg) with both oils (Fig. 1). For lavender oil, there was a significant negative effect of dose on upwind distance traveled by larvae between the 0.1- and 100-mg doses ( $r^2 = 0.49, F = 152.9$ , df = 149, P < 0.001). For pennyroyal oil, there was also a significant negative effect of oil dose on upwind distance traveled by larvae between the 0.1- and 100-mg doses ( $r^2 = 0.09, F = 20.6, df = 199, P < 0.001$ ). Forward movement of larvae in the olfactometer was significantly reduced relative to the corresponding control both by lavender oil at the 1-mg (t = 7.0, df = 39, P < 0.001, 10 mg (t = 9.6, df = 39, P < 0.001), and100 mg (t = 16.2, df = 39, P < 0.001) doses, and by pennyroyal oil at the 0.1-mg (t = 2.2, df = 49, P =0.032, 10-mg (t = 5.7, df = 49, P < 0.001), and 100-mg (t = 9.5, df = 49, P < 0.001) doses.

In the olfactometer, an effect of aging (time of exposure) of a lavender oil application was evident (Fig. 2). However, the mean upwind distance attained by larvae in the olfactometer with lavender oil application was significantly reduced compared with controls 1 h (t = 10.4, df = 29, P < 0.001), 3 h (t = 8.1, df = 29, P < 0.001), 7 h (t = 7.7, df = 29, P < 0.001), and 24 h (t = 2.1, df = 29, P = 0.04) following application of the oil to a filter paper. Mean upwind distance attained by larvae in the olfactometer 24 h after the treatment was applied was significantly less than the mean distance attained 1, 3, or 7 h after treatment, by the Tukey test at P < 0.05% (following an ANOVA; df = 119, F = 14.5).

**Barrier Tests.** The 27 essential oils varied greatly in effectiveness in the barrier assay (Tables 2 and 3), ranging from nearly no effect to strong repellency in preventing codling moth larvae from crossing the barrier in the 1-h time frame of the test. In this assay,

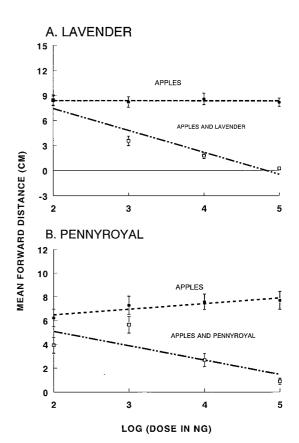


Fig. 1. (A) Mean  $\pm$  SE upwind distance attained (in centimeters) by neonate codling moth larvae in 2 min in a parallel tube olfactometer in response to air from over 3 thinning apples and oil of lavender ( $\Box$ ), or 3 thinning apples and no oil of lavender ( $\Box$ ), or 3 thinning apples and no oil of lavender ( $\odot$ ), for lavender doses from 0.1 to 100 mg, y = 12.7-2.63x log (x), where y = distance in centimeters and x = dose of oil. (B) Mean upwind distance attained (in centimeters) by neonate codling moth larvae in 2 min in a parallel tube olfactometer in response to air from over 3 thinning apples and oil of pennyroyal ( $\Box$ ), or 3 thinning apples and no oil of pennyroyal ( $\odot$ ), for pennyroyal doses from 0.1 to 100 mg, y = 7.5-1.2x log (x), where y = distance in cm and x = dose of oil.

effectiveness was indicated by a significant prolonging of the time required by larvae to cross the barrier and reach an apple. By this measure, 20 of the 27 plant oils tested were repellent to codling moth larvae (P < 0.05; Table 2). Oils of rue, garlic, tansy, and patchouly were selected for further testing based on the greatly increased times for larvae crossing these oils. As in the olfactometer assay, several plant oils provided no effect on larval behavior in the barrier assay, most notably coriander, *Coriandrum sativum* L.; wintergreen, *Gautheria procumbens* L.; spearmint, *Mentha spicata* L.; and Lavendin-S, *Lavendula X intermedia*.

Repellency was directly indicated by significant numbers of turn-arounds by codling moth larvae at the barriers (Table 3). Turn-arounds were noted each time a larva approached a barrier and reversed the direction of movement away from the oil. These

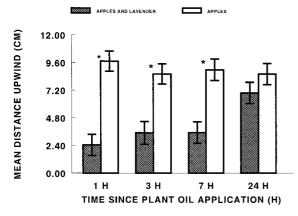


Fig. 2. Mean upwind distance attained by neonate codling moth larvae in 2 min in a parallel tube olfactometer in response to air from over 3 thinning apples and a 10-mg dosage of oil of lavender applied to a filter paper 1, 3, 7, or 24 h before the onset of bioassays. Means for treatment (crosshatched bars) and control (open bars) within a treatment age category that are significantly different (*t*-test,  $P \le 0.05$ ) are indicated by an asterisk.

counts were pooled for each set of 5 larvae. Significant increases in numbers of turnarounds were noted with 8 essential oils including rue, garlic, and tansy. Numbers of turn-arounds for dill, wintergreen, spearmint, and Lavendin-S were nearly identical to that of the experimental controls (application of solvent only).

Table 2. Mean times for neonate codling moth larvae to cross 1 of 2 plant essential oil barriers on a glass rod and to reach a thinning apple

Plant	Mean $\pm$ SE	Paire	Paired t-test results <sup>a</sup>			
riant	Treatment	Control	t	Р	df	
Rue	$30.1\pm6.9$	$1.6\pm0.1$	4.18	0.001	14	
Garlic	$18.8 \pm 3.3$	$1.5 \pm 0.1$	5.37	< 0.001	14	
Tansy	$17.1 \pm 3.1$	$2.2 \pm 0.3$	5.15	< 0.001	14	
Patchouly	$15.6 \pm 2.2$	$2.0 \pm 0.2$	6.29	< 0.001	14	
Bitter orange	$12.9 \pm 0.5$	$3.1 \pm 0.3$	5.71	< 0.001	14	
Balsam fir	$12.9 \pm 1.9$	$3.1 \pm 0.6$	5.71	< 0.001	14	
Pennyroyal	$11.3 \pm 2.4$	$3.3 \pm 0.4$	3.58	0.003	14	
Basil	$10.3 \pm 1.6$	$2.4 \pm 0.3$	4.05	0.001	14	
Eucalyptus	$10.1\pm1.6$	$2.4 \pm 0.3$	5.97	< 0.001	14	
Tagetes	$7.8 \pm 1.6$	$1.8 \pm 0.2$	4.12	0.001	14	
Sage	$7.6 \pm 1.4$	$1.6 \pm 0.2$	4.53	0.001	14	
Cypress	$7.5 \pm 1.8$	$2.4 \pm 0.3$	3.05	0.009	14	
Ginger	$6.9\pm0.7$	$3.3 \pm 0.4$	6.39	< 0.001	14	
Peppermint	$5.3 \pm 0.7$	$1.6 \pm 0.2$	5.48	< 0.001	14	
Lavender	$5.0 \pm 0.8$	$1.3 \pm 0.1$	5.43	< 0.001	14	
Spruce	$4.7\pm0.6$	$3.1 \pm 0.6$	3.42	0.004	14	
Citronella	$4.0 \pm 0.6$	$3.6 \pm 0.6$	1.28	0.220	14	
Spearmint	$3.9 \pm 0.5$	$3.1 \pm 0.4$	0.33	0.740	13	
Grapefruit	$3.6\pm0.6$	$3.3 \pm 0.4$	0.98	0.350	13	
Juniper	$3.7 \pm 0.4$	$2.5 \pm 0.3$	4.74	0.001	14	
Scotch pine	$3.5 \pm 0.5$	$1.7 \pm 0.2$	5.48	< 0.001	14	
Lavendin-S	$3.4 \pm 0.5$	$3.1 \pm 0.4$	0.33	0.740	13	
Lemon	$3.2 \pm 0.5$	$2.2 \pm 0.1$	4.15	0.001	14	
Wintergreen	$3.2 \pm 0.4$	$3.1 \pm 0.6$	0.04	0.970	14	
Dill	$3.1 \pm 0.4$	$1.7 \pm 0.2$	4.72	0.001	14	
Coriander	$2.5\pm0.4$	$2.2 \pm 0.1$	1.06	0.310	14	
Rosemary	$2.2\pm0.3$	$2.9\pm0.6$	2.15	0.050	14	

<sup>a</sup> Results from paired *t*-tests comparing responses for treatment versus control.

Table 3. Mean numbers of turn-arounds for 5 neonate codling moth larvae at either of 2 plant essential oil barriers on a glass rod

Plant	Mean no. turn-		Paired <i>t</i> -test results <sup><i>a</i></sup>		
	Treatment	Control	t	Р	
Ginger	$5.0\pm0.0$	$0\pm 0$	>13	< 0.001	
Garlic	$18.3 \pm 2.3$	$0\pm 0$	7.86	0.016	
Eucalyptus	$7.7 \pm 1.2$	$0\pm 0$	6.40	0.024	
Lavender	$4.7\pm0.9$	$0\pm 0$	5.29	0.030	
Tagetes	$6.7 \pm 1.2$	$0 \pm 0.$	5.55	0.031	
Rue	$10.1 \pm 1.7$	$0\pm 0$	5.00	0.037	
Lemon	$1.7 \pm 0.3$	$0\pm 0$	5.00	0.037	
Tansy	$4.7 \pm 1.5$	$0.3\pm0.3$	6.00	0.040	
Sage	$7.3 \pm 1.7$	$0.3 \pm 0.3$	3.50	0.073	
Citronella	$2.0 \pm 0.6$	$0\pm 0$	3.46	0.074	
Patchouly	$12.0 \pm 3.5$	$0\pm 0$	3.42	0.080	
Pennyroyal	$5.7 \pm 2.0$	$0\pm 0$	2.79	0.110	
Balsam fir	$11.7 \pm 4.7$	$0.3 \pm 0.3$	2.59	0.120	
Basil	$18.3 \pm 3.3$	$0.3 \pm 0.3$	2.67	0.120	
Peppermint	$4.3 \pm 2.3$	$0\pm 0$	1.86	0.200	
Grapefruit	$1.0 \pm 0.6$	$0\pm 0$	1.73	0.220	
Spruce	$2.3\pm0.9$	$0.3\pm0.3$	1.73	0.230	
Cypress	$7.0 \pm 4.6$	$0.3 \pm 0.3$	1.41	0.290	
Scotch pine	$2.0 \pm 1.5$	$0\pm 0$	1.31	0.320	
Spearmint	$0.3\pm0.3$	$0\pm 0$	1.00	0.420	
Lavendin-S	$0.3 \pm 0.3$	$0\pm 0$	1.00	0.420	
Bitter orange	$0.3 \pm 0.3$	$0\pm 0$	1.0	0.420	
Wintergreen	$0.0\pm0.0$	$0.3\pm0.3$	_	_	
Dill	$0.3 \pm 0.3$	$0\pm 0$	1.00	0.420	
Coriander	$0.7\pm0.7$	$0\pm 0$	1.00	0.420	
Rosemary	$0.0\pm0.0$	$0\pm 0$	—	_	

 $^a$  Results are from paired t-tests comparing responses for treatment versus control, df = 2.

Regression analyses showed that the mean times for larvae to cross a barrier increased significantly with the increased concentration of oil applied to the glass rod (Table 4), for garlic, patchouly, rue, and tansy oils. Also, the numbers of times larvae turned around at barriers increased with increasing concentration of oils applied to the glass rods for rue, tansy, garlic, and patchouly (Table 5). At the highest concentration tested in the barrier assay (100 mg/ml), most larvae had not crossed the barrier at the end of the assay (1 h).

#### Discussion

The results of these experiments indicate that several plant essential oils are promising as arrestants and repellents against larval codling moth. In the olfactometer, the odors from a number of plant essential oils greatly reduced upwind progress of neonate larvae, most notably lavender and pennyroyal. In the barrier assay, larval progress toward apples was severely impeded by rue, garlic, tansy, and patchouly oils. Although additional plant essential oils probably also are suitably effective against the codling moth, these 4 were selected for further experimentation.

The chemical constituents of these oils that are responsible for the observed effects against codling moth are unknown. Isolation and identification of these compounds may be desirable to determine the costs and efficacy of using synthetic chemicals rather than essential oils as larval arrestants or repellents.

Plant oil	Mean	Mean $\pm$ SE time (min) to cross plant oil for various concn of oil (mg/ml)				Statistics <sup>a</sup>		
	0 mg/ml	0.1 mg/ml	1  mg/ml	10  mg/ml	100 mg/ml	$r^2$	F	Р
Garlic	$2.5\pm0.2$	$2.2 \pm 0.2$	$6.2 \pm 0.6$	$44.3 \pm 3.5$	$60.0 \pm 0.0$	0.80	392.7	< 0.001
Tansy	$2.9 \pm 0.4$	$3.4 \pm 0.6$	$3.3 \pm 0.3$	$24.5 \pm 3.6$	$60.0 \pm 0.0$	0.74	273.1	< 0.001
Rue	$2.4 \pm 0.2$	$2.5 \pm 0.2$	$2.8 \pm 0.4$	$28.0 \pm 4.5$	$42.3 \pm 4.2$	0.51	37.1	< 0.001
Patchouly	$3.6 \pm 1.4$	$3.5\pm0.5$	$5.6\pm0.5$	$56.3 \pm 15.7$	$60.0\pm0.0$	0.23	37.1	0.001

Table 4. Time required for neonate codling moth larvae to cross plant oil applied to a glass rod in a laboratory assay, at different concentrations of various plant oils and statistics for the time versus concentration responses

 $^{a}$  df = 99 for all plant oils.

These essential oils were selected in part based on their reported activity against insects (Jacobson 1990). Cypress is resistant to insect attack, possibly because of deterrent and repellent chemistry. Lavender and pennyroyal are repellent to a broad variety of insects, as is patchouly, which is also insecticidal. Rue is repellent to adult Japanese beetles, *Popillia japonica* Newman. None of these oils have previously been reported to be repellent to the codling moth.

Although we refer here to arrest and repellency responses in these assays, the effects of these plant essential oils on the orientation behavior of the codling moth larvae are not entirely understood. In the olfactometer tests, reduced upwind movement in comparison with the control was documented in response to odors from the plant oils in the glass tubes. Although labeled as arrest following the terminology of Dethier (1960), this could be the result of a number of other effects, including masking of attractive odors from apple or repellency. In the barrier assay, larvae repeatedly contacted the plant essential oil barriers and turned away. This appeared to be a negative chemotropic response that can be clearly referred to as repellency. Larvae in the barrier assay also may have been slowed or confused by plant oil treatments. For the purposes of these studies, our primary interest is in preventing larval arrival at apple fruit, which is well documented here.

Differences in relative performance of the oils in the 2 assays indicate likely differences in the nature of the chemicals responsible for the arrest or repellency responses we observed. Plant oils that ranked highest in the olfactometer (lavender and pennyroyal) were not the highest ranking in the barrier test (Table 2) and vice versa. Indeed, rue and patchouly performed exceedingly well in the barrier test and did not provide significant arrest in the olfactometer. Chemical activity in the olfactometer is dependent on the release of

suitable amounts of volatile compounds from the solution of plant oil applied to filter paper, whereas the effects on behavior observed in the barrier test may be caused in part by less volatile compounds detected by larvae upon contact with the residue of oil.

There are no other studies of plant extracts or oils as arrestants or repellents of codling moth larval orientation to apple. Studies of plant extract effects on lepidopterous larvae and other insects generally involve deterrence of feeding or mortality (Jacobson 1990), with no evidence of effects on orientation or movement.

These results provide a set of candidate materials for future investigation to protect apple fruit from attack by codling moth larvae. Conceivably, plant oils may be applied directly to apple foliage and near fruit to confuse, arrest, or repel codling moth. Dispensers releasing volatile plant odorants may be used to protect trees or individual fruit. Additional experimentation needs to be done on apple trees in the field to evaluate plant oil efficacy in preventing infestation of apple by neonate larvae. Studies also need to be conducted to evaluate the same plant oils as oviposition deterrents and orientation disruptants against adult codling moths. Ideally, application of such materials would work best if they are effective against both adults and neonate larvae of the codling moth.

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Table 5. Number of times that 5 neonate codling moth larvae turned around at plant oil barriers on glass rod in a laboratory assay, at different concentrations of the plant oil applied, and statistics for turning responses versus concentration of oil

Plant oil	Mean ± SE no. times that 5 larvae turned around at the plant oil barrier for various concn (mg/ml) of plant oil applied					Reg	Regression statistics <sup>a</sup>		
	0 mg/ml	0.1 mg/ml	1  mg/ml	10  mg/ml	100 mg/ml	$r^2$	F	Р	
Garlic	$0.0 \pm 0.0$	$0.0 \pm 0.0$	$4.0 \pm 0.9$	$22.0 \pm 1.6$	$25.2 \pm 2.0$	0.82	103.0	< 0.001	
Tansy	$0.0 \pm 0.0$	$0.2 \pm 0.2$	$0.0\pm0.0$	$10.2 \pm 1.8$	$18.2 \pm 2.7$	0.72	59.1	< 0.001	
Rue	$0.0 \pm 0.0$	$0.4 \pm 0.2$	$0.6 \pm 04$	$10.8 \pm 1.2$	$11.8 \pm 1.5$	0.69	50.1	< 0.001	
Patchouly	$0.0\pm0.0$	$0.0 \pm 9.0$	$2.8\pm0.7$	$11.2\pm3.5$	$16.0\pm4.7$	0.54	26.6	< 0.001	

 $^{a}$  df = 24 for all plant oils.

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