Male Oriental Fruit Moth Response to a Combined Pheromone-Based Attracticide Formulation Targeting Both Oriental Fruit Moth and Codling Moth (Lepidoptera: Tortricidae)

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ABSTRACT Combined attracticide formulations targeting Oriental fruit moth, *Grapholita molesta* (Busck), and codling moth, *Cydia pomonella* (L.), were tested in a field trapping experiment. Capture of male codling moths in traps baited with the combined formulation was reduced compared with traps baited with the codling moth formulation alone, whereas capture of male Oriental fruit moth was increased compared with traps baited with the Oriental fruit moth formulation alone. Subsequent wind tunnel experiments showed that a single locus of the mixed attracticide formulation or close parallel presentation of the two formulations enhanced source contact by male Oriental fruit moths but did not influence earlier behaviors. However, the two formulations presented in a serial arrangement to Oriental fruit moth males in the wind tunnel resulted in enhanced lock-on, upwind flight, and source contact behaviors. In addition, male Oriental fruit moths remained on mixed pheromone droplets of the paste matrix longer than on droplets of the Oriental fruit moth formulation alone. The increased time spent on the mixed droplet was correlated with a more rapid poisoning and a greater proportion of poisoned males compared with males exposed to the Oriental fruit moth attracticide alone. These results demonstrate that a combined attracticide formulation will have different effects on each of the targeted species. It is anticipated that, due to decreased attractiveness, a combined formulation would be less effective against the codling moth. However, a mixed formulation, due to increased attractiveness and toxicity, could be more effective against the Oriental fruit moth under field conditions.

KEY WORDS pheromone, attract and kill, attracticide, mating disruption, lure and kill

THE TWO KEY PESTS OF tree fruit production in most temperate fruit-growing regions of the world are both species in the subfamily Oleuthreutinae (Lepidoptera: Tortricidae): codling moth, *Cydia pomonella* (L.), and Oriental fruit moth, *Grapholita molesta* (Busck). Larvae are internal feeders of primarily pome and stone fruits, respectively (Hill 1987). However, infestation by the Oriental fruit moth in pome fruit orchards has been on the increase in recent years in many fruit-growing regions around the world (Dorn et al. 2001), including in the northeastern United States (Usmani and Shearer 2001). In the northeastern United States, adults of both species occur in the same orchards where their flights overlap late in the season (M.L.E., unpublished data). This host expansion by the Oriental fruit moth presents an opportunity to investigate combined management strategies against these two important pests. The reliance of both of these species on a female-produced sex pheromone for mate attraction (Oriental fruit moth: Roelofs et al. 1969, Cardé et al. 1979; codling moth: Roelofs et al. 1971, El-Sayed et al. 1999) makes their chemical communication system an ideal target for integrated pest management.

Mating disruption has been developed and is commercially available for both Oriental fruit moth (Rice and Kirsch 1990) and codling moth (Charmillot 1990); however, a combined formulation has not yet been developed. More recently, an attracticide formulation that consists of a viscous paste that incorporates insecticide and sex pheromone in a UV-blocking carrier material has been developed (Hofer and Brassel 1992). This product is registered in the United States for use against Oriental fruit moth and codling moth under the trade names LastCallOFM and LastCallCM, respectively (IPM Tech Inc., Portland, OR). Success of LastCallCM in field trials (Charmillot et al. 2000, Krupke et al. 2002) and preliminary laboratory (Evenden and McLaughlin 2004a) and field (Evenden and McLaughlin 2004b) experimentation on LastCallOFM suggests it is a technology that could be effective against these tortricids on a large scale.

A great strength and potential shortcoming of pheromone-based control tactics, including mating disruption and attracticide formulations, is their specificity

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to only one or a few target pests. Some mating disruption programs have attempted to control multiple pest species with common pheromone components (van Deventer and Blommers, 1992, Pfeiffer et al., 1993, Deland et al., 1994) or a combination of pheromone components and pheromone antagonists that act interspecifically (Bengtsson et al., 1994, Evenden et al. 1999a). However, mating disruption can be achieved by several different mechanisms (Bartell 1982, Cardé 1990) and formulations do not necessarily need to be attractive to be effective (Evenden et al. 1999a, b). In contrast, the effectiveness of an attracticide depends, at least in part, on insect exposure to insecticide through source contact with the formulation (Charmillot et al. 1996, Suckling and Brockerhoff 1999) and therefore needs to be highly attractive.

There are several difficulties with dispensing more than one species' pheromone from a single formulation. For the formulation to be attractive to more than one species, a combined formulation needs to deliver the correct amount and component ratio for each species' pheromone (Weatherston 1990). Perhaps more importantly, it is crucial to determine that interspecific pheromone components do not act as antagonists to upwind flight, source contact or close range behaviors in the targeted species. Heterospecific pheromone components, which inhibit pheromonal response of other species, can be found in both closely and distantly related lepidopteran species that occur sympatrically and synchronically. Little is known about interspecific communication among oleuthreutines (Lepidoptera: Tortricidae) that overlap sympatrically and synchronically. Male lesser apple moth, *Grapholita prunivora* (Walsh) (Lepidoptera: Tortricidae), response to pheromone-baited traps is significantly reduced with incorporation of a component of Oriental fruit moth pheromone, (Z)-8-dodecenol (Baker and Cardé 1979). This compound seems to be important in the reproductive isolation of these closely related species (Baker and Cardé 1979). One compound, dodecanol (12:OH) has been reported from the effluvia of both codling moth (Arn et al. 1985, Bäckman et al. 1997) and Oriental fruit moth (Cardé et al. 1979). However, neither of the LastCall formulations that we tested contained this compound. LastCallCM contains only the major pheromone component of the codling moth, (*E*8-*E*10)-dodecadienol (codlemone) (Roelofs et al. 1971), and LastCallOFM contains a three-component blend of (*Z*)-8-dodecenyl acetate (*Z*8–12:Ac), (*E*)-8-dodecenyl acetate (*E*8–12:Ac), and (*Z*)-8-dodecenol (*Z*8–12:OH) (Cardé et al. 1979). Incorporation of the major component of the Oriental fruit moth pheromone, *Z*8–12:Ac (Roelofs et al. 1969), into cylindrical traps baited with synthetic sources of codlemone or virgin female codling moths resulted in a reduction of the number of male codling moths captured (Arn et al. 1974). The other pheromone components of the Oriental fruit moth, *Z*8–12:OH and *E*8–12:Ac (Cardé et al. 1979), also inhibited male codling moth capture when added individually to traps baited with synthetic sources of codlemone (Arn et al. 1974). Incorporation of *Z*8–12:OH into a source of codlemone inhibited upwind flight of male codling moths in a wind tunnel (Preiss and Priesner 1988).

Here, we tested the impact of combining the two LastCall formulations on the attraction of both species in the field. In addition, we examined upwind flight behavior and toxicity of the formulations to male Oriental fruit moths in a wind tunnel. Our experiments were designed to examine the impact of a combined formulation in presentation scenarios that males might encounter these formulations in an orchard setting. A completely integrated formulation targeting the two species would result in single droplets containing both species' pheromones. Alternatively, a single dispenser could house the two formulations separately and have two tips so that droplets are applied together but separated by a certain distance resulting in distinct plumes close to the source. Finally, a single orchard could be treated individually with separate formulations creating the possibility of serial and parallel overlapping plumes.

Materials and Methods

LastCall Formulations. LastCall formulations used in field and wind tunnel studies were formulated by D. Czokajlo (IPM Tech Inc.). Formulations consisted of a clear viscous paste with a base of a proprietary product plus other inert ingredients (93.8% of the formulation). Oriental fruit moth pheromone used in the LastCallOFM formulation was purchased as a premixed three component blend that consisted of 90–94.5% *Z*8–12:Ac, 5.5–7.2% *E*8–12:Ac, and 0.75–1.5% *Z*8–12:OH with each component >97% pure (Bedoukian Research Inc., Danbury, CT). Headspace analysis of the LastCallOFM formulation used in these experiments was conducted by R. Gries (Department. Biological Sciences, Simon Fraser University, Burnaby, British Columbia, Canada) and confirmed the release of a 100:7.4:1.54 ratio of *Z*8–12:Ac: *E*8–12:Ac and *Z*8–12:OH after a 1-d aeration of 10, 50-mg droplets of material. The *Z*8–12:Ac:*Z*8–12:OH ratio released changed to 100:4.3 after a 2-d aeration. Codling moth pheromone used in the LastCallCM formulation consisted of 100% codlemone that was 97% pure (Ciba-Geigy, Basel, Switzerland). In each formulation, the pheromone was incorporated into the LastCall formulation at 0.16% with 6.0% of the insecticide permethrin. Individual LastCall sources consisted of a 50-μl droplet positioned in the center of a 1 by 2-cm piece of aluminum foil and suspended vertically by a short length of wire in the trap or wind tunnel. For field use, droplets were prepared the afternoon before travel to field sites. Foil pieces with droplets were suspended individually within glass jars and held in the refrigerator until transport to field sites within a refrigerated container. LastCall droplets used in the wind tunnel were weighed to between 42.5 and 52.5 mg and used in bioassays within 1 h of preparation.

Field Trapping Experiment. A trapping experiment was conducted in five commercial orchards in southeastern Pennsylvania during the 2003 field season. Four Intercept A traps (IPM Tech Inc.) were positioned 15 m apart in apple trees, 1.5 m above the ground. Traps were baited with foil tabs containing one of four treatments: 1) a single 50-μl droplet of LastCallOFM; 2) a single 50-μl droplet of LastCallOFM and a single 50-μl droplet of LastCallCM separated by 1 cm; 3) a 50-μl droplet of LastCallOFM and a 50-μl droplet of LastCallCM placed in the center of the foil tab and mixed together; and 4) a 50-μl droplet of LastCallCM. One of the four treatments was assigned to each trap randomly, and male moth catches were counted and removed after 1 wk. In total, eight, 1-wk trapping periods were conducted throughout the field season. At each trapping period, trap position within the orchard was randomized and new LastCall droplets were suspended in traps. Male moth catches for each species in traps baited with one of the four treatments were log $(x + 1)$ transformed and compared using a randomized block design analysis of variance (ANOVA) with orchard specified as a random factor and trapping period treated as a repeated measure (PROC MIXED, SAS Institute 1996). When no moths were captured in any of the baited traps at a given orchard, that orchard was eliminated from analysis for that trapping period. Analysis of Variance was followed by Least Square Means tests (SAS Institute 1996) within each species to assess male attraction to individual treatments.

Insects. Male moths used in wind tunnel bioassays came from an Oriental fruit moth laboratory colony maintained on a lima bean-based diet at a photoperiod of 16:8 (L:D) h and 24°C. Pupae were separated by sex and males were held under a reversed photoregime until moth eclosion and subsequent use in the bioassay. Adult males were held in individual clear, 30-ml cups and provided with a water source. Two-5-d-old males were collected 40 min before testing and placed under bioassay conditions in the wind tunnel room.

Wind Tunnel. The flight section of the wind tunnel was 1.7 m in length and 0.85 m in height. Air was pushed through the tunnel at speeds between 0.4 and 0.55 m/s, and the pheromone plume was exhausted by a centrally located fan at the downwind end. Uniform dim white light was provided by six, 25-Watt incandescent bulbs, diffused through white paper. Temperature was maintained between 20 and 25°C in all bioassays.

Wind Tunnel Experiments. Wind tunnel bioassays were conducted during the last 2 h of the photophase and the first hour of the scotophase. LastCall formulations were presented to male moths on foil tabs suspended from a bar extended from a ring stand, 40 cm off the floor and 10 cm from the upwind end of the tunnel. Males were transferred individually in conical screen release cages (5 cm in diameter by 5 cm in height) just before introduction at the downwind end of the wind tunnel on a platform 40 cm off the floor and 20 cm from the downwind end of the tunnel. Male response to the various semiochemical treatments was graded as + or − for wing fanning, locking-on to the plume, oriented upwind flight, and source contact. Males had 2 min to respond to the pheromone source or they were considered nonresponsive.

The first wind tunnel experiment, experiment 1, tested the hypothesis that the addition of LastCallCM to LastCallOFM would influence male Oriental fruit moth upwind flight response in a wind tunnel. Treatments tested were 1) a single 50-μl droplet of LastCallOFM, 2) a 50-μl droplet of LastCallOFM and a 50-μl droplet of LastCallCM placed in the center of the foil tab and mixed together, and 3) a single 50-μl droplet of LastCallOFM and a single 50-μl droplet of LastCallCM dispensed on two separate foil tabs and separated horizontally by 2 cm along the bar to produce two distinct plumes close to the source. This separation was chosen as smoke tests illustrated separate plumes leaving the release devices at this distance. For the third treatment, position of the droplets (left or right) was assigned randomly on each day of testing, and moths were only marked + for source contact if contact with the LastCallOFM droplet occurred. Ten to 12 males were presented individually to each treatment on each of 7 d for a total of 85 moths flown to each treatment. The proportion of males that conducted each behavior in the wind tunnel was compared using logistic regression (PROC LOGISTIC, SAS Institute 1996). The contrast statement in PROC LOGISTIC was used to compare any significant treatment effects.

Experiment 2 compared male response to LastCallOFM when formulations were alone or presented in various upwind/downwind positions to establish serially overlapping plumes with LastCallCM. Four treatments were presented to males: 1) a single 50-μl droplet of LastCallOFM positioned in the normal upwind position, 140 cm upwind of the release device; 2) a single 50-μl droplet of LastCallOFM positioned 110 cm upwind of the release device; 3) a single 50-μl droplet of LastCallOFM positioned at 110 cm and a single 50-μl droplet of LastCallCM positioned at 140 cm upwind of the release device; and 4) a single 50-μl droplet of LastCallOFM positioned at 140 cm and a single 50-μl droplet of LastCallCM positioned at 110 cm upwind of the release device. For the third and fourth treatments, moths were only marked + for source contact if contact with the LastCallOFM droplet occurred. Ten males were presented individually to each treatment on each of 8 d for a total of 80 moths flown to each treatment. Logistic regression was used to analyze this two-factor treatment: LastCallOFM alone or serially combined with LastCallCM; and position of LastCallOFM droplet: upwind or downwind) factorial experiment for each of the behaviors conducted (SAS Institute 1996).

Toxicity Experiment. Experiment 3 tested the hypothesis that the addition of LastCallCM to LastCallOFM would influence the toxicity of the formulation to male Oriental fruit moths. To expose males to the formulations in a way most similar to a field situation, we conducted another wind tunnel experiment that compared only 1) a single 50-μl droplet of LastCallOFM; and 2) a 50-μl droplet of LastCallOFM and a 50-μl droplet of LastCallCM placed in the center of the foil tab and mixed together. Nine to 15 males were presented individually to each treatment on each of 9 d for a total of 125 moths flown to each treatment. This experiment was assessed in three ways: 1) behaviorally, as in experiments 1 and 2; 2) the time spent on the droplet was recorded using a handheld stopwatch for males that contacted the droplet; and 3) toxicity of the formulations to males that contacted the droplet assessed 3, 24, and 48 h postexposure. Males that contacted the droplet in the wind tunnel were captured using a small net and placed in a 30-ml cup. All males were transported to the laboratory, provided with a water source, and arranged in random order on a tray left in full light for the duration of the experiment. Males were checked at 3, 24, and 48 h postexposure and rated as healthy, incapacitated, or dead. The proportions of males that conducted the various behaviors in the wind tunnel to the two treatments were compared by logistic regression (PROC LOGISTIC, SAS Institute 1996). The times spent on the droplet were compared using a randomized block ANOVA with replicate and treatment specified as explanatory variables (PROC GLM, SAS Institute 1996). Males that got stuck in the viscous droplet (n = 3) were removed from the analysis. The proportion of healthy, incapacitated and dead males was compared for each treatment at each assessment period by logistic regression (PROC LOGISTIC, SAS Institute 1996).

Results

Capture of codling moth males in the field trapping experiment was greatest in traps baited with LastCallCM alone and was significantly reduced by the addition of LastCallOFM either as a mixture or with droplets of the two formulations separated by 1 cm. No male codling moths were attracted to the LastCallOFM formulation alone (Fig. 1). Conversely, although capture of male Oriental fruit moths was extremely low in the trapping experiment, it was significantly increased by the combination of formulations in either of the two formats. As with the codling moth response, no male Oriental fruit moths were captured in traps baited with the heterospecific formulation alone (Fig. 1).

In the first wind tunnel experiment, there was no significant difference in the proportion of Oriental fruit moth males that wing-fanned, took-off from the release device, locked-on to the semiochemical plume, and conducted oriented upwind flight to the LastCallOFM droplet alone or in combination with LastCallCM in either presentation format. However, significantly more males contacted the LastCallOFM droplets that were combined with or presented in parallel with LastCallCM than contacted LastCallOFM droplets alone (Fig. 2).

In experiment 2, the proportion of males that locked-on and conducted upwind oriented flight to the LastCallOFM droplet was significantly increased by an overlapping LastCallCM plume placed either up- or downwind in a serial presentation (Fig. 3).

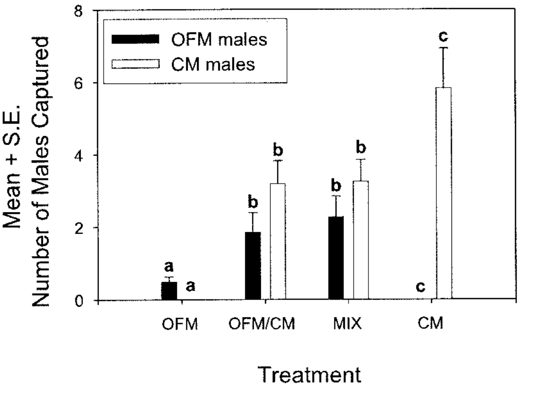

Fig. 1. Mean + SE males captured in traps baited with 50-μl droplets of LastCallOFM and LastCallCM alone and in combination (OFM/CM: droplets of each formulation separated by 1 cm; MIX: droplets of each combination thoroughly combined). N = 8. Within each species, bars with the same letter are not significantly different, Least Square Means ($P > 0.05$).

Removal of nonsignificant position and interaction effects from the model revealed a marginally significant treatment effect (P = 0.0538) on the proportion of males that contacted the LastCallOFM droplets (Fig. 3). There was no significant position effect or position by treatment interaction effect on the proportion of males that conducted all behaviors in the wind tunnel in experiment 2.

Behaviorally, the responses of Oriental fruit moth males in experiment 3 were consistent with the first wind tunnel experiment. There was no difference in the proportion of males that conducted the initial behaviors to either the LastCallOFM droplet alone or to the mixed droplet and again a significantly greater proportion of males contacted the droplet that contained the mixture of the two formulations. In addition, males that contacted the droplets in experiment 3 stayed for a significantly longer period of time on the droplets that contained both formulations (Fig. 4). Finally, the toxicity to males exposed to the two treatments in experiment 3 varied as well (Fig. 5). There was a significant treatment effect due to the formulation that males were exposed to on the proportion of males ranked as healthy at 3, 24, and 48 h postexposure (P = 0.0004). However, there was no time since exposure effect or time*treatment effect on males that were ranked as healthy. In comparison, there were significant treatment ($P < 0.0001$), time since exposure and time*treatment ($P < 0.0001$) effects on the proportion of males that were ranked as incapacitated. This suggests that the overall level of poisoning was greater and the rate of poisoning was faster for males exposed to the mixed formulation. Finally, there was no treatment effect on the proportion of males that were ranked as dead at 3, 24, and 48 h (P = 0.2738), but there was a significant time*treatment effect (P = 0.0007). All males exposed to the mixed formulation were dead at 48 h postexposure compared with only

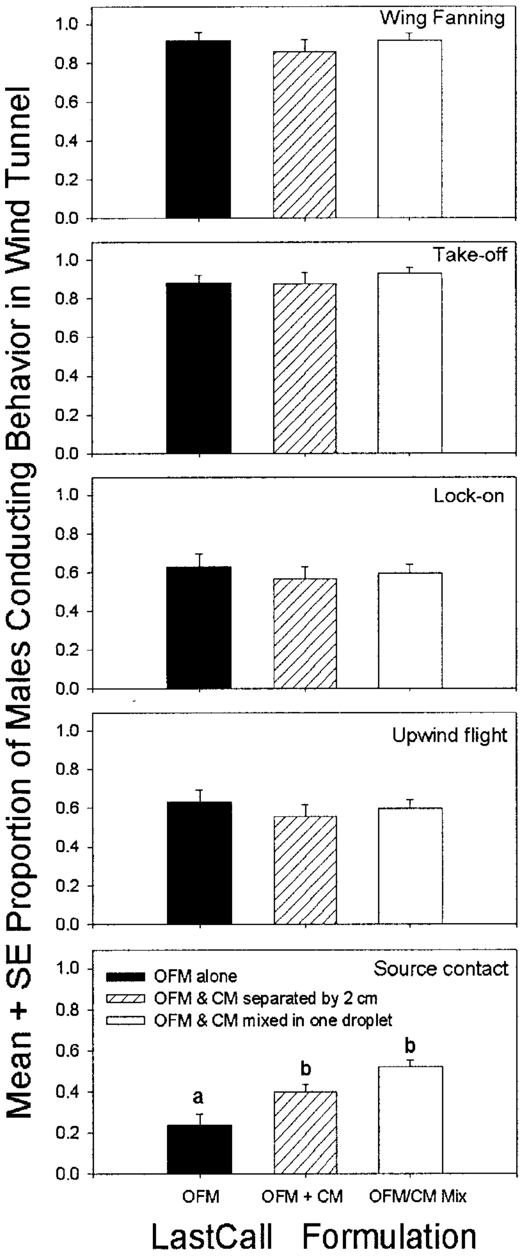

Fig. 2. Mean + SE proportions of male Oriental fruit moth exhibiting behavioral responses to a 50-μl droplet of LastCallOFM alone, presented in parallel with a LastCallCM droplet and completely mixed with a LastCallCM droplet in the wind tunnel. Bars with the same letter are not significantly different, logistic regression ($P > 0.05$).

76.7% of the males exposed to the LastCallOFM formulation alone (Fig. 5).

Discussion

Our study provides mixed results on the potential for the development of a combined attracticide formulation to target codling moth and Oriental fruit moth. The field trapping experiment demonstrated that male codling moth response to LastCallCM droplets was inhibited by the incorporation of the LastCallOFM formulation either as a mixture or as two distinct droplets separated by 1 cm. Inhibition of pheromone response by male codling moths to sources containing the Oriental fruit moth pheromone is not surprising. Trap capture was significantly reduced by incorporation of individual Oriental fruit moth pheromone components into codlemone-baited traps (Arn et al. 1974).

Our data demonstrate that there was no significant difference in the trap-capture reduction of male codling moth to the two formulations presented as a completely integrated droplet or separated by 1 cm. Most studies on behavioral antagonists of pheromonal response in the Lepidoptera have demonstrated that simultaneous perception of the antagonist and the pheromone is required for inhibition of male upwind flight response (Witzgall and Priesner 1991, Liu and Haynes 1992, Rumbo et al. 1993, Fadamiro and Baker 1997) because male moths are able to discriminate between individual pheromone filaments (Mafra-Neto and Cardé 1994, Vickers and Baker 1994). In our trapping study, a 1-cm separation of the two droplets probably resulted in significant mixing of the pheromone filaments released and resulted in simultaneous perception of the two sources. Similar results were found in a trapping study of cabbage looper moth, *Trichoplusia ni* (Hübner) (Lepidoptera: Noctuidae). Behavioral antagonists that were released from separate sources within a trap baited with the *T. ni* pheromone significantly reduced male moth capture compared with traps baited with sex pheromone alone (Mitchell 1976). The reduction of male codling moth response to the combined formulation would be detrimental to the efficacy of a combined attracticide formulation as fewer codling moth males would touch the droplets and be removed from the population. However, only 50% of the efficacy of LastCallCM tested alone in large-scale field trials was attributed to removal of poisoned male moths from the population (Charmillot et al. 1996), suggesting that source contact with the droplets is not necessary to obtain some level of control.

Although very low numbers of male Oriental fruit moths were captured in the trapping experiment, a significantly greater trap catch occurred in traps baited with the combined formulation in either of the two presentation formats. It is unusual to have pheromone response enhanced or synergized by the incorporation of a heterospecific pheromone component. However, our data demonstrate that the LastCallOFM formulation alone is not an optimally attractive lure. This same formulation was not as attractive as calling virgin females tested in a wind tunnel in previous work (Evenden and McLaughlin 2004a). It may be that the addition of codlemone in the LastCallCM formulation simply served to compensate for the suboptimal level of Z8–12:OH in the Oriental fruit moth formulation. Alternatively, the release rate

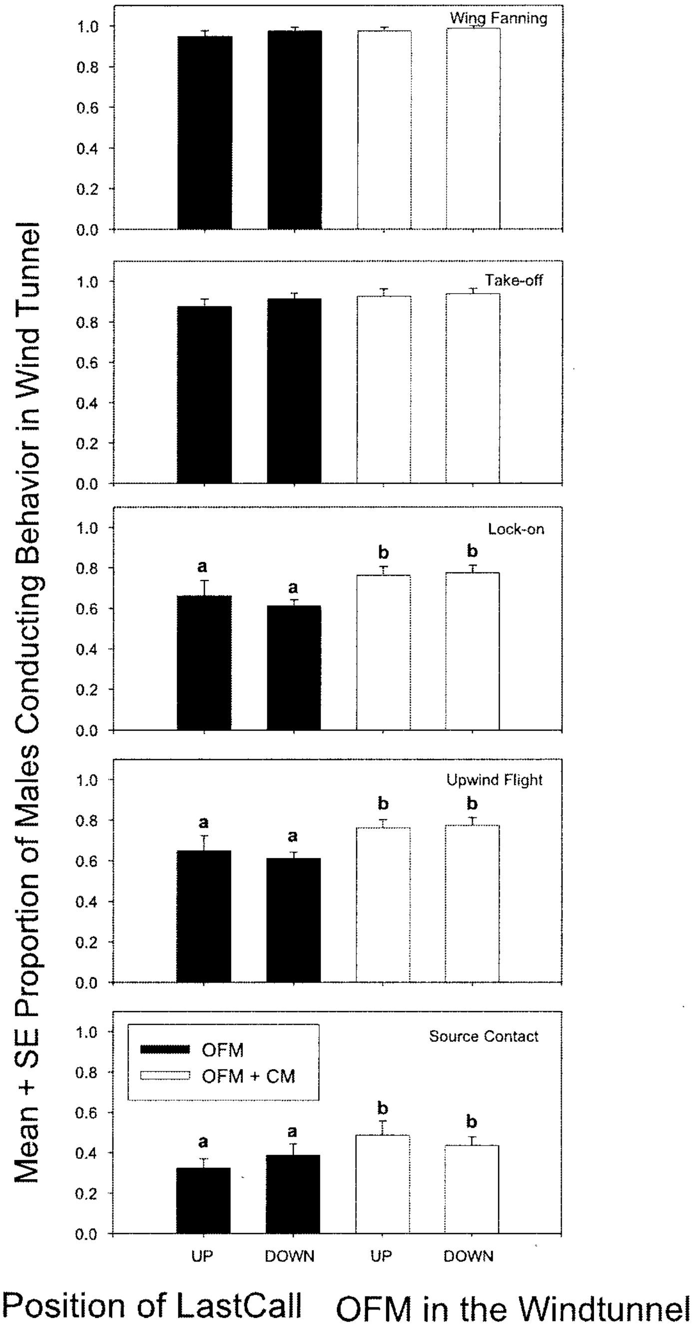

Fig. 3. Mean + SE proportions of male Oriental fruit moth exhibiting behavioral responses to a 50-μl droplet of LastCallOFM alone and presented in a serial arrangement with a 50-μl droplet of LastCallCM in the wind tunnel. Up refers to the LastCallOFM droplet in the upwind position (140 cm from the release platform). Down refers to the LastCallOFM droplet in the downwind position (110 cm from the release platform). Bars with the same letter are not significantly different, logistic regression ($P > 0.05$).

from the LastCallOFM formulation may be too high to elicit optimal response. However, other researchers working with different commercial pheromone formulations for these species (L. Hull and P. Shearer, personal communications) also have observed synergism in male Oriental fruit moth response to combined formulations. Compounds acting as pheromone synergists to tortricid moths (Roelofs and Comeau 1971), including the Oriental fruit moth (Roelofs et al. 1973), have been recorded. Large amounts of dodecanol

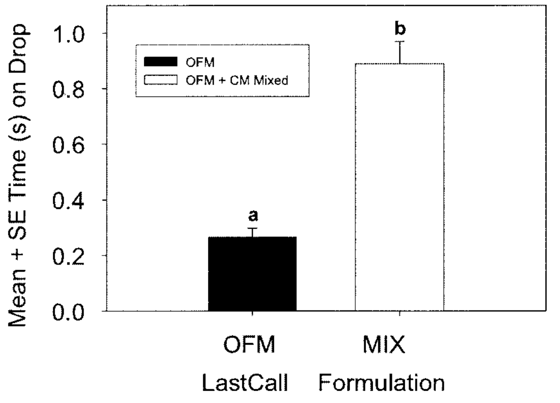

Fig. 4. Mean + SE time spent by male Oriental fruit moths on a 50-μl droplet of LastCallOFM alone or combined with a LastCallCM droplet in the wind tunnel. Bars with the same letter are not significantly different, ANOVA ($P >$ 0.05).

(12:OH) and 8-propoxyoctan-1-ol also synergized male Oriental fruit moth capture in traps baited with a suboptimal blend containing the major pheromone component Z8–12:Ac (Roelofs et al. 1973) with ≈6% *E*8–12:Ac (Linn et al. 1986). However, 12:OH was later identified in the effluvia of Oriental fruit moth females (Cardé et al. 1979) and did not enhance upwind flight when released at an appropriate ratio within the full pheromone blend (Baker and Cardé 1979). It remains to be seen if the addition of codlemone to an optimal Oriental fruit moth pheromone blend containing ≈10% Z8–12:OH will enhance male response.

Closer observation of male Oriental fruit moths in the wind tunnel in experiments 1–3, enabled us to determine what aspects of upwind flight behavior were impacted by the combination of the two formulations in the various presentation formats. When the two formulations were completely integrated in one droplet in experiments 1 and 3, only source contact was significantly increased as a result of the addition of LastCallCM to LastCallOFM (Fig. 2). However, a greater proportion of male Oriental fruit moth males locked-on to the pheromone plume and conducted upwind oriented flight and source contact when presented with overlapping plumes of the two formulations in experiment 2 (Fig. 3). Close-range behaviors at the droplet also were enhanced as evidenced by an increased time spent on the droplets containing the mixed formulation (Fig. 4). Our findings are similar to early studies that showed incorporation of high ratios of 12:OH into a suboptimal Oriental fruit moth pheromone blend consisting only of the two acetates (Cardé et al. 1975) or with reduced amounts of the important Z8–12:OH component (Baker and Cardé 1979) resulted in an increase in the frequency of close range behaviors, including hairpencil display. However, incorporation of 12:OH at appropriate ratios in the full pheromone blend exhibited no effect on male behavior (Baker and Cardé 1979). In our study, codlemone, also a 12-carbon alcohol, was released at a high ≈1:1 ratio with the Oriental fruit moth blend in the mixed formulations and therefore may be perceived by antennal receptors and impact male Oriental fruit moth behavior in a similar manner to high release ratios of 12:OH in combination with the suboptimal pheromone blend. Interestingly, male codling moth attraction to codlemone in a wind tunnel is enhanced by the addition of host plant volatiles including 12:OH (Coracini et al. 2004). Another possible explanation for the observed synergism in our study is that codlemone is mimicking a host plant volatile that enhances Oriental fruit moth upwind flight behavior to its pheromone.

The increased time spent on the droplets that contained the mixed formulation (Fig. 4) resulted in a more rapid poisoning of males and a greater proportion of poisoned males (Fig. 5). The rate of poisoning of males exposed to the LastCallOFM formulation alone was similar to that observed in previous experimentation using this same formulation (Evenden and McLaughlin 2004a). The mortality rates at 24 h postexposure in both experiments are similar to the 56.8% mortality rate of male pinkbollworm moths, *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelechiidae), flown to an attracticide formulation containing 10% permethrin in a wind tunnel (Haynes et al. 1986). In the current study, at the final assessment conducted 48 h after exposure, there was still 20% of the males that touched the LastCallOFM droplet that were healthy, whereas almost all of the remaining males were dead (Fig. 5). This suggests that moths graded as healthy 48 h postexposure would not be affected by the permethrin. In contrast, all of the males that contacted the mixed droplets were dead 48 h postexposure.

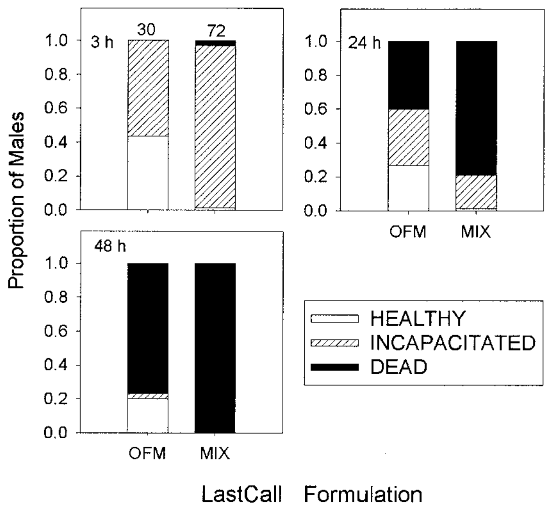

Fig. 5. Toxicity of LastCallOFM alone and combined with LastCallCM to male Oriental fruit moths, 3, 24 and 48 h postexposure in the wind tunnel.

Our results have revealed that a combination of the two LastCall formulations will have different effects on each of the targeted species. A combined formulation inhibited male codling moth attraction and enhanced male Oriental fruit moth attraction to the attracticide. However, some male codling moths were captured in traps baited with the combination and so a mixed formulation might work well in orchards that have a minor codling moth problem and higher Oriental fruit moth populations. Oriental fruit moth males contacted mixed droplets more readily and remained on the mixed droplets for a longer period of time resulting in overall increased effectiveness of the mixed formulation. Therefore, unless the LastCallOFM formulation is changed in the future to release more of the important pheromone component Z8–12:OH, the combination of LastCall formulations has the potential to be just as effective or more effective than LastCallOFM alone.

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