Towards Developing Colorado Potato Beetle Attracticide: Importance of Inert Ingredients

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Abstract

Viscous formulation of the kairomone blend based on potato plant volatiles showed in a number of recent studies a potential for aggregating Colorado potato beetles, *Leptinotarsa decemlineata* (Say) in treated areas of the field. However, we observed a rather unexpected decline in beetle populations on plots in small-plot field trials conducted in 2004. Follow-up field and laboratory studies conducted in Maine and Maryland determined that inert carrier used in the formulation was toxic to beetle larvae. These findings are important for interpreting results of future field trials involving this particular formulation because observed effects would inevitably result from an interaction between the kairomone and its carrier, not from the kairomone alone.

Introduction

A recently synthesized kairomone blend, based on the volatiles produced by potato plants, has been demonstrated to be attractive to both adult and larval stages of the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Dickens 1999, 2000, 2002). It was subsequently formulated in a viscous inert carrier for field applications and showed potential for aggregating beetles in treated areas of the field (Martel et al. 2005 a, b). This opens a possibility to manipulate beetle behavior for control purposes. However, in our small-plot field trials conducted in 2003 in Maine (Alyokhin et al. 2004), less frequent (compared to Martel et al. [2005a, b]) applications resulted in a rather unexpected decline in beetle populations on plots treated with the kairomone formulation. The objective of this study was to determine if the observed effect was consistent between the years, and whether reduction in beetle populations observed during the previous season was due to the kairomone, or due to its inert carrier.

Materials and Methods

Sampling procedures. For each treatment, six 17.7 m long and 4 row wide experimental plots were planted to certified seed potatoes ('Kennebec'), providing us with five replications per treatment. Plots were arranged in a randomized complete block design. Approximately 1.8 m was left between the plots within each block, and blocks were spaced at ~3 m. For the treatments receiving kairomone formulation or the inert carrier, applications on three plots at each treatment were made using commercially available paint sprayers. Applications on the other three plots at the same treatment were made using disposable plastic syringes. Kairomone formulation and the inert carrier were applied at a rate of 0.9 ml per 1 row meter (Martel et al. 2005a), but the application technique was different from the one used by Martel et al. (2005a). Twenty plants were randomly selected at weekly intervals from each plot. The number of Colorado potato beetles in all life stages on each selected plant was recorded.

Field Experiment. The objective of this experiment was to determine whether reduction in beetle populations observed during the previous season was due to the kairomone formulation, or due to its inert carrier. Experiments were replicated at the Aroostook Research Farm in Presque Isle, Maine, and at the USDA-ARS Research Facility in Beltsville, Maryland during the 2004 growing season. We compared beetle densities and defoliation levels on control plots, plots treated with the kairomone formulation, and plots treated with blank inert carrier. Beetle densities were determined by weekly visual counts. Defoliation levels were visually estimated throughout growing season in Maryland and between 28 July and 19 August in Maine, where they were too small to detect earlier in the season. Kairomone formulation and and carrier were applied twice during the season. The first spray was directed against overwintering adults and was applied at 80% plant emergence from the soil. The second spray was directed against small larvae. The data were analyzed using repeated measures ANOVA.

Laboratory Experiment. The objective of this experiment was to test toxicity of kairomone, blank matrix, and attracticide (kairomone+inert carrier+spinosad) to the Colorado potato beetle larvae under controlled laboratory conditions. Formulations were applied in the amount of 0, 5, or 50 mg (in 0, 2, or 10 droplets respectively) to two potato leaves placed in a distilled water pic, then housed in plastic ventilated 300 ml Nalgene containers. Ten newly hatched 1st instar larvae from the IBL lab culture (field origin Maryland) were placed on the foliage not contacting the kairomone drops. Containers were housed in growth chambers at 25°C and 16-hour photoperiod with RH ~50%. The larvae were checked at 1, 2, 4, 5, 6, and 8 days after the start of the experiment, with number of live and dead larvae, and their instar, noted at each date. Foliage was replaced when more than 70% defoliated, but treated leaves were left in the container. Results were evaluated by single-degree-of-freedom pairwise comparisons on each day using Fisher's Exact test with a Bonferroni correction for an overall experimentwise α=0.05 on each day.

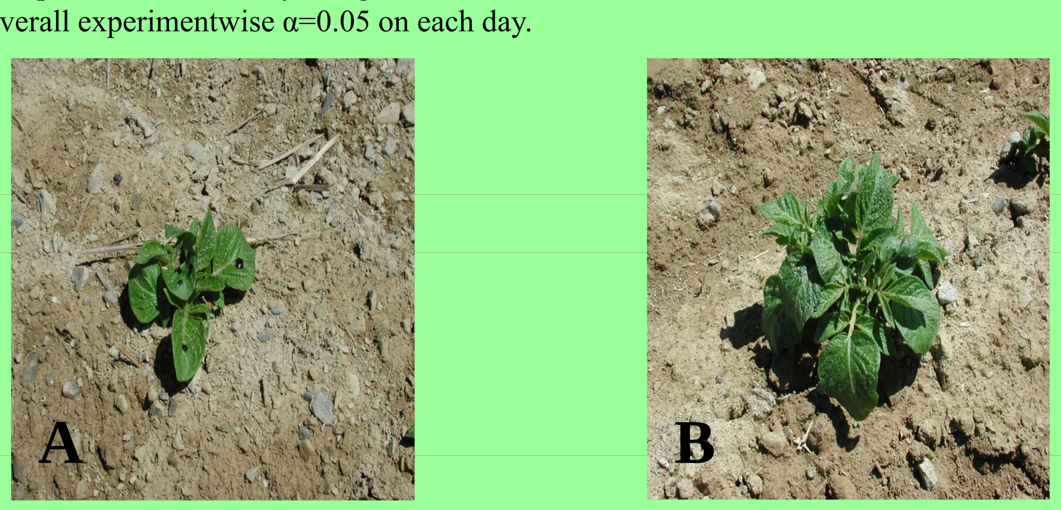


Fig. 1. Kairomone formulation (black globs) applied to the newly emerged potato plots. (A) Aplications made using syringes. (B) Applications made using paint sprayer.

Results

Field Experiment. In Maine, applications of both kairomone and blank matrix significantly reduced the populations of small larvae and adult beetles (Fig. 2A). There was no statistically significant difference in large larvae. Defoliation followed the same pattern as large larvae (Fig. 3A) and also was not statistically significant. In Maryland, beetle densities were quite variable (Fig. 2B) and did not appear to follow any particular pattern. None of treatment effects were statistically significant. The same applied to defoliation ratings (Fig. 3B).

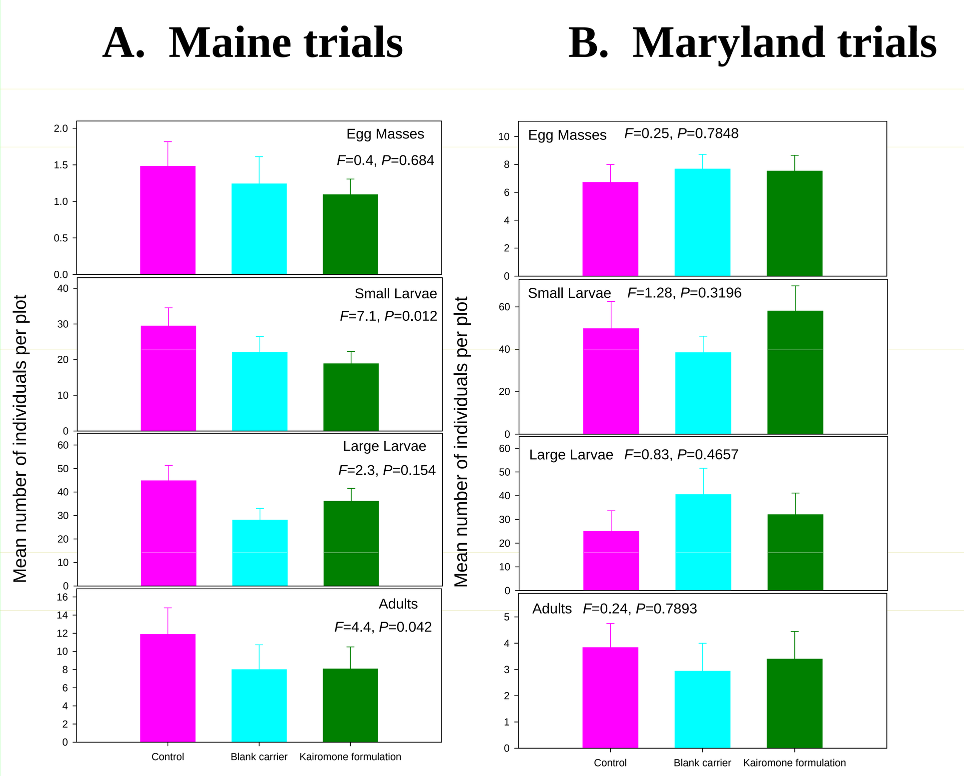
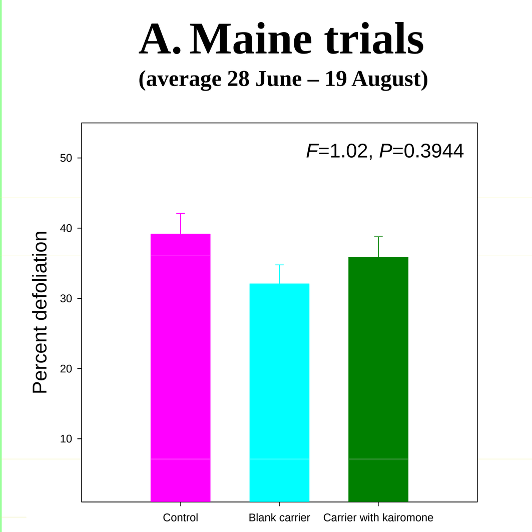


Fig.2. Colorado potato beetle densities on plots treated with blank carrier and kairomone formulation (carrier + kairomone).



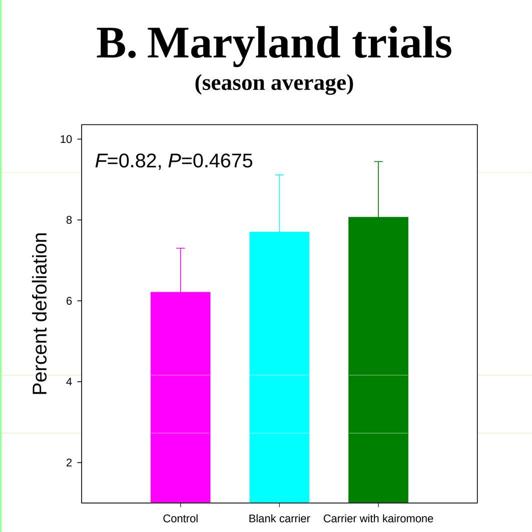


Fig. 3. Defoliation readings on plots treated with blank carrier and kairomone formulation (carrier + kairomone).

In Maine, densities of all beetle life stages (Fig. 4A), as well as defoliation ratings (Fig. 5A) were numerically lower on the plots treated with drop applications. Except for adults, the differences were statistically significant only at α=0.1. In Maryland, the pattern was not very consistent, and the effect of the application method was not statistically significant (Figs. 4B and 5B).

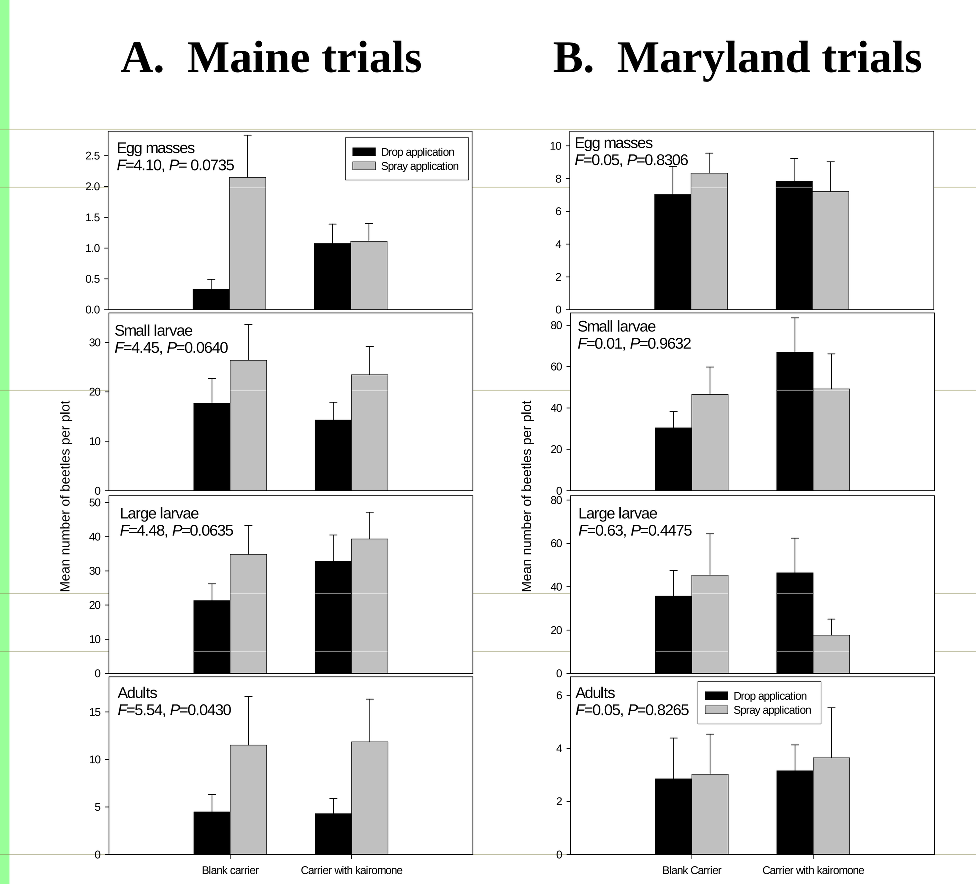
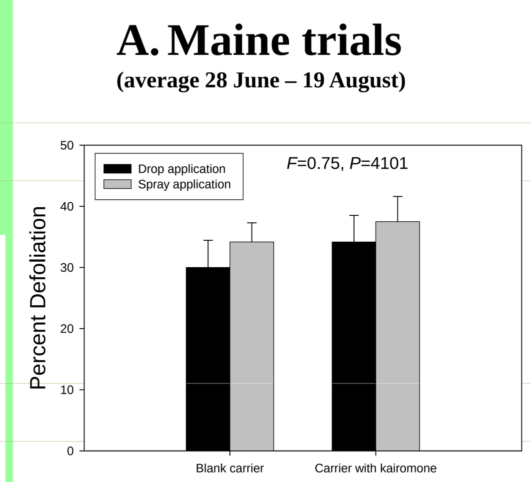


Fig. 4. Colorado potato beetle densities on plots treated using disposable plastic syringes (drop applications) or paint sprayer (spray applications).



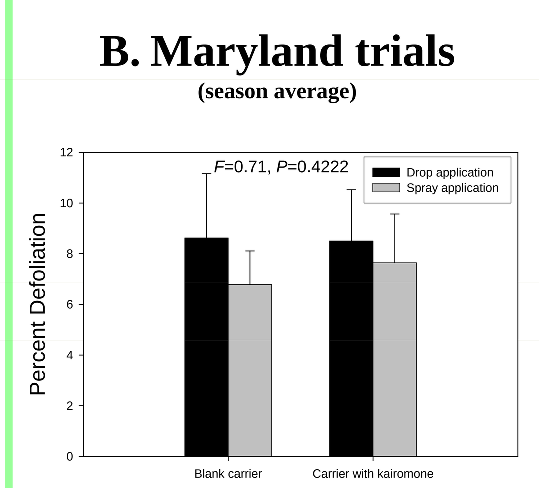


Fig. 5. Defoliation readings on plots treated with blank carrier and kairomone formulation (carrier + kairomone).

Results (continued)

Spinosad-containing attracticide, kairomone, and blank matrix significantly increased beetle mortality (Table 1; Fig. 6). Spinosad-containing formulations produced significant mortality compared to control at first inspection, day 1. Kairomone and blank formulations showed significant mortality compared to control only beginning on day 4. There was no significant difference in response in kairomone and blank treatments on any date. However, bait formulations showed increased mortality compared to kairomone-only formulations on all dates. There was also a significant dose response for all formulation considered as a group, beginning with day 2.

Fig. 6. Survival of neonate Colorado potato beetle larvae exposed to attracticide, kairomone, and blank matrix .

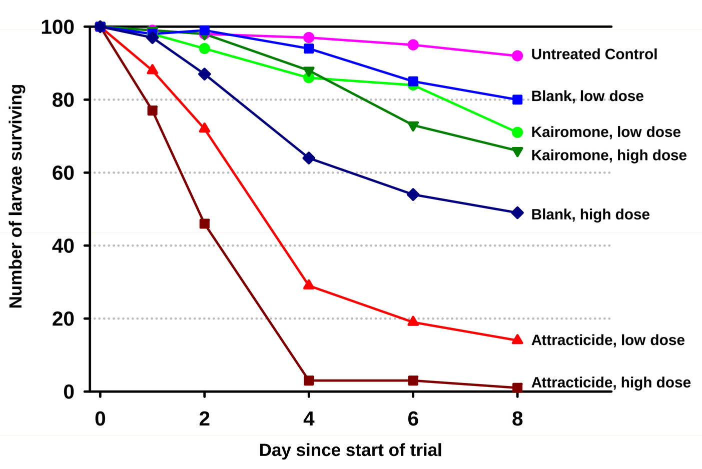


Table 1. Results of multiple comparisons between treatments in the laboratory experiments. "YES" indicates significantly more larvae survived in first-mentioned group at day shown. "NO" indicates no significant difference. See text for details on experimental design and statistical tests involved.

	DAY 1	DAY 2	DAY 4	DAY 6	DAY 8
Low vs. High dose (all formulations combined)	NO	YES	YES	YES	YES
Control vs. Inert carrier	NO	NO	YES	YES	YES
Control vs. Kairomone	NO	NO	YES	YES	YES
Control vs. Attracticide	YES	YES	YES	YES	YES
Inert carrier vs. Kairomone	NO	NO	NO	NO	NO
Kairomone vs. Attracticide	YES	YES	YES	YES	YES

Discussion

Our results indicate that inert carrier used to formulate Colorado potato beetle kairomone has a negative effect on the Colorado potato beetle larvae. This effect is not very strong. Although rather pronounced in the laboratory, it was somewhat masked in the field, presumably by other variables affecting beetle populations. In Maine, where population densities were relatively low, there was a fairly consistent trend towards lower beetle numbers on plots treated with either kairomone formulation, or blank inert carrier. In Maryland, where beetle densities were four times as high as in Maine, the effect was not discernable by our sampling methods. It is possible that at higher beetle densities other mortality factors became more important in controlling beetle populations, and relatively weak effect of carrier substance disappeared in background variation.

It is likely that physical contact with the carrier was detrimental for beetle larvae. In laboratory studies, more droplets applied to potato leaflets resulted in higher mortality. Also, mixing kairomone formulation with spinosad resulted in a dramatic increase in larval mortality. Since spinosad was limited to the droplets of formulation, the larvae had to come in contact with them to become poisoned. Also, in Maine trials lower beetle densities were found on plots treated using syringes, the technique that produced larger and more persistent droplets of treatment materials. It should be noted that volatile collections from treated plants after exposure in the field for one week revealed only minute amounts each component of the three component blend from plants treated with syringe droplets, while an essential component of the attractant was absent from volatile collections from plants treated with the paint sprayer (Alford, Alyokhin, Weber and Dickens, unpublished). Since the interval between treatments in the current study was several weeks the contribution, if any, of the kairomone odor to the effects noted in the current study is questionable.

These findings have important implications for developing attractants that might be used to manipulate Colorado potato beetle behavior to improve beetle control. By itself, higher beetle mortality induced by the kairomone carrier may be desirable. However, it should be kept in mind when interpreting results of field trials involving this particular formulation because observed effects would inevitably result from an interaction between the kairomone and its carrier, not from the kairomone alone.

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