Effects of naturally occurring and synthetic synergists on the toxicity of insecticides and xanthotoxin to *Amyelois transitella* (Lepidoptera: Pyralidae)

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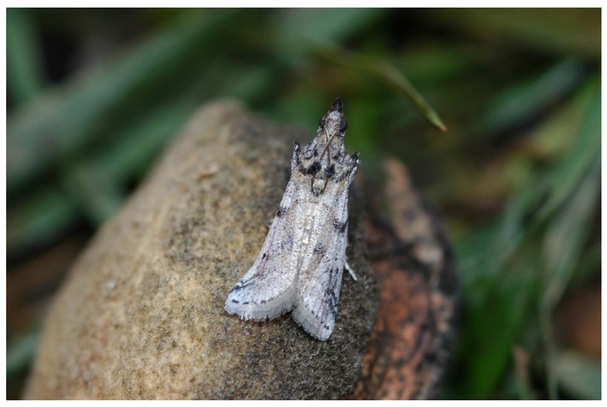
Introduction

Infested almond



Adult navel orangeworm

The navel orangeworm, *Amyelois transitella*, is among the most destructive pests of almonds in California, as well as a serious problem in pistachios (*Pistacia vera)*, figs (*Ficus carica*) and walnuts (*Juglans regia*). Neonates tunnel into the nut and successive instars consume the nutmeat, generating large quantities of frass and webbing. In addition to causing such direct losses, the insect feeding leaves almonds vulnerable to infection by *Aspergillus* spp. that produce toxic aflatoxins (Campbell et al., 2003). The adults lay eggs in mummy fruits when new crop nuts are unavailable and the caterpillars have been reported as scavengers on mummified fruits or nuts of at least 25 plant species.

Fungal contamination in orchards is frequently reported to be associated with the insect damage and the control of the insect decreases aflatoxin levels in crops. Currently, the management depends on a combination of control tactics, including cultural practices and insecticide sprays, particularly when infested adjacent crops, such as pistachios, provide immigrants into almond orchards. Biological control agents, although identified, cannot yet provide effective control. Chemical sprays include organophosphates and pyrethroids; at present for hull splits which expose nuts to insect invasion and hull rot fungi when the hull, the outside fleshy material covering the shell of the nut, splits. Only Entrust (spinosad) sprays are acceptable in organic orchards.

Some naturally occurring essential oil constituents, such as myristicin, are known synergists for certain synthetic insecticides (Lichtenstein and Casida, 1963) and phytochemicals, such as xanthotoxin (Berenbaum and Neal, 1985). Piperonyl butoxide (PBO), a well-known synthetic inhibitor of P450s with similar structure of MYR has been widely reported to synergize the toxicity of the pyrethroid insecticides (Ishaaya, 1993). In this study, we set out to ascertain the ability of essential oil constituents to synergize the toxicity of host plant phytochemicals and insecticides and subsequently reduce survival of navel ornageworms.

Objectives:

1. Determine the toxicity of insecticides to *Amyelois transitella* in laboratory bioassays;
2. Determine the efficacy of natural and synthetic synergists on insecticide toxicity.

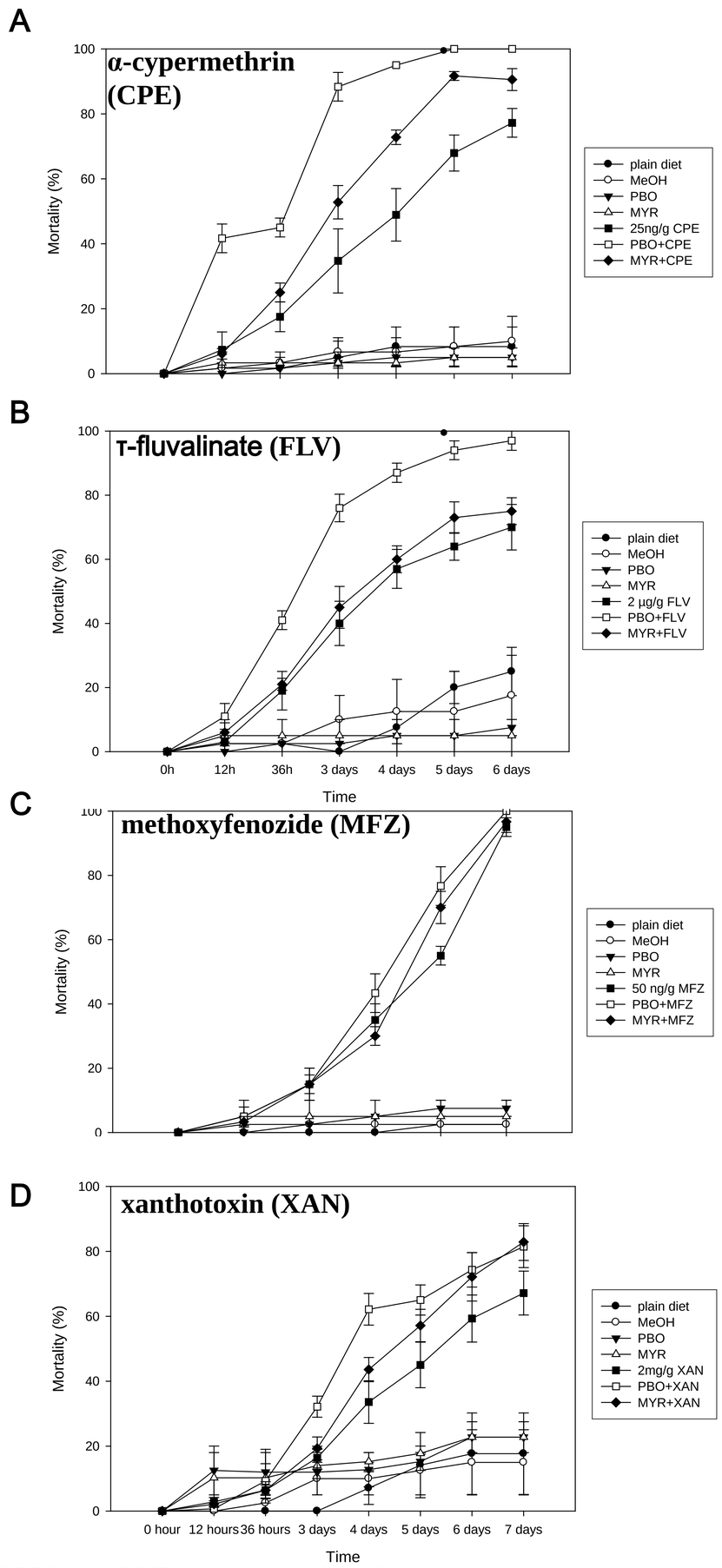
Results

1. LC50 of insecticides for first instar *Amyelois transitella*

Insecticides	24h LC_{50} (95% Confidence Limits) µg/g	48h LC_{50} (95% Confidence Limits) µg/g
τ-fluvalinate	1.9 (1.2-3.0)	0.73 (0.45-1.2)
α-cypermethrin	0.072(0.045-0.11)	0.013 (0.007-0.023)
methoxyfenozide	-	0.076 (0.043-0.13)

Mortality of first instars treated with varying concentrations of insecticides was recorded after 24 and 48 hours. All bioassays were repeated at least three times and the LC50 (the concentration of the insecticide in the diets that causes 50% mortality at the specified time) was calculated with the Probit analysis function from SPSS v17 software (SPSS Inc., Chicago, IL).

2. Bioassays of potential synergists including myristicin (MYR) and Piperonyl butoxide (PBO) of insecticide and xanthotoxin toxicity to first instar *Amyelois transitella*.



Effects of PBO and MYR on the mortality of newly hatched larvae exposed to cypermethrin (CPE) (A), fluvalinate (FLV) (B), methoxyfenozide (MFZ) (C) and xanthotoxin (XAN) (D) were tested. Larvae were fed with artificial diets containing 25 ng/g CPE, 2 µg/g FLV, 50 ng/g MFZ or 2 mg/g XAN supplemented with or without 25 µg/g MYR or 200 µg/g PBO. Plain diet and diets containing the synergists alone or an equal amount of MeOH were used as controls.

Conclusions:

1. Piperonyl butoxide significantly synergizes cypermethrin, fluvalinate and xanthotoxin and increases their toxicity to the larvae,;
2. Myristicin synergizes cypermethrin and only slightly increases mortality of the larvae fed with xanthotoxin after 7 days;
3. Myristicin can be explored as a field treatment to reduce survival of this pest species and aflatoxin contamination in orchard situations.

Acknowledgments

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References

Campbell, B. C., R. J. Molyneux, and T. F. Schatzki. 2003. Current research on reducing pre- and post-harvest aflatoxin contamination of US almond, pistachio, and walnut. Journal of Toxicology Toxin Reviews 22: 225-66. **Lichtenstein, E. P., and J. E. Casida. 1963.** Myristicin, an insecticide and synergist occurring naturally in the edible parts of parsnips. J Agric Food Chem. 11: 410–5. **Berenbaum, M., and J. J. Neal. 1985.** Synergism between myristicin and xanthotoxin, a naturally co-occurring plant toxicant. J Chem Ecol. 11: 1349–58.