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Lethal and sublethal effects of seven insecticides on three beneficial insects in laboratory assays and field trials

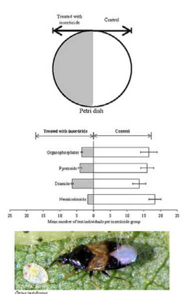
Maria E.S. Fernandes [a], Flávia M. Alves [b], Renata C. Pereira [c], Leonardo A. Aquino [a], Flávio L. Fernandes [a, *], José C. Zanuncio [d]

[a] Universidade Federal de Viçosa, Campus Rio Paranaíba, Instituto de Ciências Agrárias, Rio Paranaíba, MG 38810-000, Brazil
[b] Universidade Federal de Viçosa, Campus Viçosa, Departamento de Fitotecnia, Viçosa, MG 36570-000, Brazil
[c] Universidade Federal de Viçosa, Campus Viçosa, Agroecologia, Viçosa, MG 36570-000, Brazil
[d] Universidade Federal de Viçosa, Campus Viçosa, Departamento de Entomologia, Viçosa, MG 36570-000, Brazil

HIGHLIGHTS

- Safety of seven pesticides to the three beneficial insects was evaluated.
- The chlorantraniliprole showed low lethal and sublethal effects to test-species.
- Deltamethrin induced hormesis in *Cycloneda sanguinea* and *Orius insidiosus.*
- The organophosphates and pyrethroids on predators IPM be evaluated with caution.

GRAPHICAL ABSTRACT



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ABSTRACT

Lethal and sublethal effects of insecticides on target and non-target arthropods are a concern of pest management programs. *Cycloneda sanguinea*, *Orius insidiosus* and *Chauliognathus flavipes* are important biological control agents for aphids, whitefly, lepidopterus eggs, thrips and mites. All three test species were subjected to a toxicity study using the insecticides acephate, bifenthrin, chlorantraniliprole, chlorpyrifos, deltamethrin, imidacloprid, and thiamethoxam. Experiments were done in the lab and field. In the laboratory we evaluated the mortality and sublethal effects of the concentration that killed 20% of the population (LC_{20}) on feeding, repellence and reproduction of the species tested. The lethal effects of these insecticides at the recommended doses was evaluated in the field. Concentration-response bioassays indicated chlorantraniliprole had the lowest toxicity, while chlorpyrifos and acephate were the most toxic. Test species exposed to filter paper surfaces treated with pyrethroids, neonicotinoids and organophosphates were repelled. On the other hand, test species were not repelled from surfaces treated with chlorantraniliprole. Chlorantraniliprole therefore seemed to be the least dangerous insecticide for these three beneficial arthropod test species.

1. Introduction

A key element of pest management programs in agroecosystems is to build an understanding of the impacts on non-target and

* Corresponding author.
E-mail address: flaviofernandes@ufv.br (F.L. Fernandes).

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beneficial insects (Desneux et al., 2007). The use of insecticides against insect-pests still prevails as one of the main pest management tools in most agricultural settings, in addition to having potential consequences for arthropod pest resurgence (Roubos et al., 2014).

Insecticides may block some physiological or biochemical processes, impacting survival, growth, development, reproduction and behavior of natural enemies of insect pests (Desneux et al., 2007; Castro et al., 2012). Even at non-lethal levels, insecticides can still influence behavior, although there have been few detailed studies concerning the potential effects of sublethal insecticide doses on the behavior of beneficial arthropods. In general, sublethal insecticides levels affect reproduction, orientation, feeding, oviposition and learning. In many cases insecticides act as repellents associated with food searching behavior. Repellency may result from contact with the host or prey treated with insecticides (Saran et al., 2014). Moreover, there is the possibility that a phenomenon known as hormesis will occur in populations of natural enemies (Guedes and Cutler, 2014). Hormesis is defined as the stimulation of organism performances that occur at low levels of exposure to agents that are normally toxic at high levels of exposure (Calabrese and Baldwin, 2001). This phenomenon has been reported for many animal-toxicant models and has often been suggested as the main mechanism for pest population resurgence (Cordeiro et al., 2013; Qu et al., 2015).

Pyrethroids and organophosphates were introduced in the mid 1980s, followed by the neonicotinoids in 1990 (Grube et al., 2011). Compared to these insecticides, the anthranilic diamides (e.g. chlorantraniliprole) were commercialized in 2006 (Lahm et al., 2009). The oldest insecticides, deltamethrin and bifenthrin (pyrethroids); imidacloprid and thiamethoxam (neonicotinoids) and chlorpyrifos and acephate (organophosphates), are reported to cause lethal and sublethal effects on natural enemies (Desneux et al., 2007; Rajak et al., 2014), but in the majority of cases the diamides showed lesser effects on this group (Brugger et al., 2000; Moscardini et al., 2015). Pyrethroids act as sodium channel modulators, organophosphates inhibit the action of the acetylcholinesterase enzyme, neonicotinoids bind and act as agonists on acetylcholine receptors (postsynaptic nicotinic acetylcholine receptor), causing paralysis that leads to death, often within a few hours and the diamides modulate the calcium channels connecting the ryanodine receptors (IRAC, 2015). In ambient conditions, the groups cited may be degraded by biotic (Zuo et al., 2015) and abiotic action (Sharma et al., 2014).

Families such as Coccinellidae and Anthocoridae have received attention because of their importance as natural enemies to some major insect pest species. Cantharidae, on the other hand, has not been well studied. *Cycloneda sanguinea* (L.) (Coleoptera: Coccinellidae) and *Orius insidiosus* (Say) (Hemiptera: Anthocoridae) are important biological control agents for aphids, whitefly, lepidopterus eggs, thrips and mites (Oliveira et al., 2005; Lucas, 2012; Yamada et al., 2016), but little is known about *Chauliognathus flavipes* Fabricius (Coleoptera: Cantharidae). Boiça Junior et al. (2004) observed that in greenhouse conditions, *C. sanguinea* adults reduced the population of *Aphis gossypii* Glöver (Hemiptera: Aphididae) by 93.5% in 2 d. In the United States, *O. insidiosus* is a significant factor in the predictable seasonal declines in *Frankliniella* spp., following population peaks and is able to suppress *F.* spp. populations in *Capsicum annuum* L. (Reitz et al., 2003). There is little information on the predation capacity of *C. flavipes* and its use in programs of biological control, but the Cantharidae family is an important predator of aphids (Berthiaume et al., 2001).

Considering the importance of the insecticides and the above predators, the objective was to (1) evaluate the effect of the insecticides chlorpyrifos, chlorantraniliprole, deltamethrin, acephate, imidacloprid, bifenthrin and thiamethoxam on mortality in laboratory and field conditions, and (2) to evaluate the effect of the sublethal doses of these insecticides on the feeding, repellence and reproductive behavior of *O. insidiosus*, *C. sanguinea* and *C. flavipes*.

2. Materials and methods

2.1. Insects

Adults of *O. insidiosus*, *C. sanguinea* and *C. flavipes* were collected from corn (*Zea mays* L.), soy (*Glycine max* L.) and tomato (*Solanum lycopersicum* L.) plantations in experimental fields at the Universidade Federal de Viçosa, Minas Gerais State, Brazil (20°45′25″S, 42°52′55″W). The capture method used soda bottles with rectangular openings (15 × 20 cm) closed with a thin organza tissue. On the inside of the bottle a paper towel was added to facilitate insect movement. The test species were captured with a simple sucker hose, blown to the inside of the bottle and transported to the laboratory to initiate breeding.

O. insidiosus adults were raised from Mendes and Bueno (2001), with adults collected from corn plants (*Z. mays*) and separated into pairs. After separation, individuals were placed on acrylic Petri dishes (15 × 2.1 cm) sealed with polyethylene film to prevent escape. Nymphs of thrips (*F.* spp.) were used as a food source and *Bidens pilosa* L. (Asteraceae) inflorescences were used as oviposition sites. The inflorescence was observed daily with a stereoscopic microscope to verify the presence of test species eggs. The test species were kept in climatic chambers at 25.00 ± 1.00 °C, 70.00 ± 10.00% relative humidity and photoperiod of 12:12 (light:dark). The inflorescences containing *O. insidiosus* eggs were removed and collected on other Petri dishes (15 × 2.1 cm) sealed with polyethylene film. To avoid egg and nymph mortality by desiccation, a cotton ball moistened with distilled water was placed inside the dish. To avoid water condensation in the insect breeding environment, only five inflorescences of *B. pilosa* were added per container. The containers were observed three times per week to add food and to moisten the cotton. The inflorescences of *B. pilosa* used as substrate for oviposition of *O. insidiosus* females were kept in the container to provide shelter to the nymphs. When the nymphs transformed into adults they were removed to perform the bioassays.

C. sanguinea adults were raised according to Oliveira et al. (2005) and *C. flavipes* adults were collected from soy plants. Adults collected from the field were individually transferred to plastic pots (250 mL), which had 1 cm^2 circular perforations to allow gas exchange with the external environment. A paper towel was added to the inside of the pots so the adults could oviposit. The adults were removed after oviposition and the eggs were placed on glass Petri dishes (15 × 2.0 cm) with a small moist cotton ball dampened with sanitary water added to prevent the eggs from drying out. After hatching, larvae were individually transferred to 500 mL plastic pots, with about 15 aphids/pot/larva/day. When the larvae pupated they were transferred to a BOD (Biochemical Oxygen Demand) germinating chamber at a constant temperature of 25.00 ± 0.10 °C and relative humidity of 75.04 ± 0.40% to hatch the adults that were then used in the bioassays.

2.2. Insecticides

Commercial formulations of seven neurotoxic insecticides available for use on soy, corn and tomato fields in Brazil were used. These were the organophosphates chlorpyrifos (Lorsban 480 emulsifiable concentrate; i.e., containing 480 g a.i. L^{-1}, Dow Agrosciences industrial Ltda, São Paulo, SP, Brazil) and acephate (Orthene 750 water-dispersible granules, i.e. containing 750 g a.i.