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# Effects of spinosad and indoxacarb on survival, development, and reproduction of the multicolored Asian lady beetle (Coleoptera: Coccinellidae)

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**Abstract**

Use of selective insecticides, such as spinosad and indoxacarb, that are more toxic to lepidopteran pests than to *Harmonia axyridis* (Pallas), could facilitate conservation of this predator in sweet corn integrated pest management (IPM). We examined the effects of spinosad and indoxacarb on survival, development, and reproduction of *H. axyridis* by spraying first instars and adult females. Treatments for the first instar assay were spinosad at 10, 25, and 50% of the field rate (FR), indoxacarb at 10% FR, and water (untreated check). We recorded survival of each life stage, developmental time to adults, and adult weight. Treatments for the adult female assay were spinosad at 50 and 100% FR, indoxacarb at 50% FR, and water (control). Each day, we recorded female survival and reproductive capacity. Indoxacarb decreased survival of first instars and adults, extended the developmental time for first instars to become adults, and reduced the fecundity of *H. axyridis* females. Spinosad decreased survival of first instars, extended the time for first instars to become adults, decreased weight gain, and reduced the fertility of *H. axyridis* females. Our results suggest that spinosad and indoxacarb may reduce *H. axyridis* population growth by affecting its survival, development, and reproduction. We also conclude that indoxacarb, when applied at 10% FR, has more lethal and sublethal effects on *H. axyridis* than spinosad applied at 10, 25 or 50% FR. The importance of sublethal effects of insecticides, as well as acute toxicity, in toxicological studies with natural enemies is discussed within the context of biological control and IPM.


*Keywords:* *Harmonia axyridis*; Conservation biological control; Sublethal effects; Insecticides; Toxicity

## 1. Introduction

Sweet corn, throughout much of the US is intensively managed with insecticides for the European corn borer, *Ostrinia nubilalis* (Hübner), and the corn earworm, *Helicoverpa zea* (Boddie) (Bartels and Hutchison, 1995; Hutchison et al., 2004). The extensive use of insecticides in crop systems, however, may cause resurgence of the primary pest, replacement by secondary pest populations (Elzen, 2001), environmental contamination (Frank et al., 1990), effects on nontarget organisms (Croft, 1990), and development of pest resistance (Brattsten et al., 1986). Therefore, alternatives to chemically intensive pest management are necessary. In light of this necessity, the incorporation of biological control offered by existing natural enemies into integrated pest management (IPM) programs in sweet corn could reduce insecticide applications, thus benefiting growers, consumers, and the environment. The multicolored Asian lady beetle, *Harmonia axyridis* (Pallas), is a common predator in sweet corn (Musser and Shelton, 2003a; Wold et al., 2001), preying on *O. nubilalis* eggs (Hoogendoorn and Heimpel, 2002; Musser and Shelton, 2003b) and on a secondary pest, the corn leaf aphid, *Rhopalosiphum maidis* Fitch (Hoogendoorn and Heimpel, 2004). Conservation of

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this predator may enhance biological control of these pests in sweet corn.

The relatively new insecticides, spinosad and indoxacarb, are effective against several lepidopteran pests (Burkness et al., 2003; Wing et al., 2000). Since the primary pests in sweet corn are lepidopterans (i.e., *O. nubilalis* and *H. zea*), use of these insecticides on this crop is expected to increase. Spinosad is a macrocyclic lactone that causes involuntary muscle contractions, tremors, and eventually paralysis of treated insects (Salgado, 1998). Indoxacarb is an oxadiazine that makes treated insects stop feeding and go into mild convulsions or permanent paralysis (Tillman et al., 2001; Wing et al., 2000).

Spinosad and indoxacarb have shown relatively low toxicity to natural enemies (Michaud and Grant, 2003; Sparks et al., 2001) and have therefore been designated as reduced-risk insecticides (Organic Materials Review Institute, 2002). Most studies that showed the toxicity of these insecticides to natural enemies, however, used acute mortality as the primary criterion for susceptibility (Galvan et al., 2005; Michaud and Grant, 2003; Miles and Dutton, 2000; Tillman and Mulrooney, 2000). Acute toxicity tests, however, detect only lethal effects of insecticides (Stark and Banks, 2003; Stark et al., 1997). Sublethal effects of insecticides that show low toxicity to natural enemies and leave many survivors after treatment cannot be fully evaluated using acute toxicity approaches (Wennergren and Stark, 2000). For example, 80% of adult parasitic wasps, *Catolaccus grandis* (Burks), survived after exposure to spinosad (Elzen et al., 2000). However, no *C. grandis* pupae developed from *Anthonomus grandis grandis* Boheman second instars treated with spinosad at only 1/4 of the rate that resulted in 80% adult *C. grandis* survival (Elzen et al., 2000).

Research on sublethal effects seeks to discover the negative, nonlethal impacts of insecticides on various life history parameters that may affect population dynamics (e.g., Stark and Banks, 2003). Insecticides have been shown to have effects on sex ratio and time to adult emergence (Vinson, 1974), fecundity and development of the female ovipositor (Lawrence, 1981), egg hatch (Stark et al., 1992), weight gain and locomotory behavior (Vincent et al., 2000), pre-oviposition period and mutations in offspring (Stark and Banks, 2003), and feeding behavior (Singh et al., 2004). By ignoring these sublethal effects, toxicological studies that evaluate only lethal effects may underestimate the negative impacts of insecticides on natural enemy populations.

Few studies have examined the sublethal effects of insecticides on Coccinellidae (e.g., Grafton-Cardwell and Gu, 2003; Liu and Stansly, 2004; Mani et al., 1997). For *H. axyridis*, various insecticides have been shown to have sublethal effects on weight gain (methoxyfenozide) (Carton et al., 2003), mobility (imidacloprid and λ-cyhalothrin) (Provost et al., 2003; Vincent et al., 2000), and fecundity (imidacloprid) (Weissenberger et al., 1997). Because of the paucity of information on sublethal effects of spinosad and indoxacarb on *H. axyridis* and the likely increase in the use of these chemistries in agriculture, there is a need for examination of their sublethal effects. The objective of this research was to evaluate the impacts of spinosad and indoxacarb on survival, development, and reproductive capacity of *H. axyridis*.

## 2. Materials and methods

### 2.1. Insects

*Harmonia axyridis* were obtained from a laboratory colony founded from adults collected during October 2003 at the Rosemount Research and Outreach Center, University of Minnesota, Rosemount, MN. Following collection, beetles were held in 1.96 L plastic dishes with ~200 beetles per dish, and maintained at 10 ± 1 °C and a photoperiod of 16:8 (L:D) h. Prior to experimentation, the dishes containing beetles were warmed to 25 ± 1 °C with a photoperiod of 16:8 (L:D) h (these rearing conditions were used throughout the rest of the studies), and the beetles were allowed to mate for 14 days. The beetles were provided an ad libitum supply of live soybean aphids, *Aphis glycines* Matsumara, and pea aphids, *Acyrthosiphon pisum* (Harris), a diet made from freeze-dried drone honey bee, *Apis mellifera* L., pupae (Okada and Matsuka, 1973), and water in 0.5 ml plastic microcentrifuge tubes plugged with cotton. After the mating period, adult females were maintained individually in plastic petri dishes (60 × 15 mm) lined with 55 mm filter paper disks. The petri dishes containing females were checked daily for oviposition. If eggs were found, the females were removed and transferred to new petri dishes (60 × 15 mm) provisioned with food and water. After egg hatch and dispersal of larvae from egg clusters (i.e., ~1 day after hatching), individuals of the $F_1$ generation were placed individually into separate plastic petri dishes (60 × 15 mm), and were reared to the desired developmental stages (i.e., first instars or adults) on a diet of freeze-dried drone *A. mellifera* pupae.

### 2.2. First instar assay

The study was conducted as a randomized complete block design for first instar *H. axyridis*. The experiment consisted of five treatments and three replications through time, with 20–25 individuals per replication. Treatments used in this study included: spinosad (SpinTor 2SC, Dow AgroSciences LLC) at 10% of the field rate (FR) [0.011 kg AI/ha (0.0094 lb AI/ac)]; spinosad at 25% FR [0.0275 kg AI/ha (0.0235 lb AI/ac)]; spinosad at 50% FR [0.055 kg AI/ha (0.047 lb AI/ac)]; indoxacarb (Avaunt WG, E.I. du Pont de Nemours and Company) at 10% FR [0.0062 kg AI/ha (0.0055 lb AI/ac)]; and an untreated check (i.e., water). Partial rates for spinosad (10, 25, and 50% FR) and indoxacarb (10% FR) were used because field rates