# LABORATORY EVALUATION OF INSECTICIDES FOR CONTROL OF THE INVASIVE *CACTOBLASTIS CACTORUM* (LEPIDOPTERA: PYRALIDAE)

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ABSTRACT

We conducted laboratory assays of nine products registered for use on ornamental plants in Florida for their ovicidal and larvicidal activity against the invasive cactus moth *Cactoblastis cactorum*. One-hundred percent mortality (or 0% survival) of 1-day-old eggs was obtained when eggstick sections were treated with cypermethrin, spinosad, or imidacloprid. These products were equally as effective when assayed against eggs that were fully embryonated (28 days old), when cladodes of *Opuntia stricta* were exposed to neonates 24 hours after dipping, or to cladodes that were dipped and stored for 30 days before exposure. When *Bacillus thuringiensis* (Dipel®) was used to prevent neonate penetration into treated cladodes of *O. stricta*, 100% mortality (or 0% survival) was recorded in the laboratory.

Key Words: insecticides, invasives, *Cactoblastis cactorum*, Lepidoptera, Pyralidae, cypermethrin, emamectin benzoate, abamectin, spinosad, azadirachtin, fenoxycarb, imidacloprid, acephate, *Bacillus thuringiensis*

RESUMEN

Nosotros realizamos unos ensayos del laboratorio de nueve productos registrados para el uso sobre plantas ornamentales en Florida para su actividad ovicida y larvacida contra la polilla invasora de cactus *Cactoblastis cactorum*. Una mortalidad de cien por ciento (o 0% sobreviviencia) de huevos de 1 dia de edad fue obtenida en secciones de grupos de huevos tratados con cypermethrin, spinosad, o imidacloprid. Estos productos fueron igualmente efectivos en ensayos contra huevos con embriones completemente desarrollados (de 28 dias de edad), cuando los cladodios de nopal de *Opuntia stricta* fueron expuestas a neonatas (larvas recien nacidas) 24 horas después ser emergidos, o a los cladodios que fueron emergidos en insecticida y almacenados por 30 dias antes de ser expuestos. Cuando *Bacillus thuringiensis* (Dipel®) fue usado para prevenir la penetración de las neonatas dentro los cladadios de *O. stricta* tratados, un mortalidad de 100% (o 0% sobrevivencia) fue registrada en el laboratorio.

*Cactoblastis cactorum* (Berg) successfully controlled several species of invasive prickly pear cacti (Cactaceae: Opuntioideae—*Opuntia*) in Australia (Dodd 1940), South Africa (Pettey 1948), and in many other parts of the world (Moran & Zimmermann 1984). In 1989 *C. cactorum* was detected in the Florida Keys (Habeck & Bennett 1990; Dickel 1991). The cactus moth may have arrived through natural dispersal from the Caribbean Islands, where it was intentionally introduced in the 1950s (Simmonds & Bennett 1990), or it may have been accidentally introduced by the nursery trade (Pemberton 1995). Nevertheless, its rapid spread along the Atlantic and Gulf Coasts has raised concerns about its unavoidable impact on native *Opuntia* cacti in the southern United States and in Mexico (Zimmermann et al. 2000). Stiling (2002) suggested that the geographical range of *C. cactorum* in Florida was expanding at an approximate rate of 50-75 km per year. However, unpublished data collected by our group suggests that the spread rate along coastal locations in the Gulf of Mexico was closer to 160 km per year during 2000-2003 (S. D. Hight, unpublished data). Given this rapid rate of geographical expansion, *C. cactorum* could arrive in Texas by the year 2007. Invasion and establishment of the cactus moth in the southwestern United States and in Mexico will have serious detrimental effects on biodiversity and stability of native desert ecosystems and on vegetable, fruit, and forage *Opuntia* industries in these areas

(Soberón et al. 2001; Zimmermann et al. 2000). Even though *C. cactorum* was deliberately introduced into South Africa to control invasive cacti, spineless *Opuntia* is still used as fodder for cattle and other livestock during times of drought. As such, livestock farmers manage their *Opuntia* plantations in order to minimize losses due to insect damage.

The biology of *C. cactorum* is well documented (Dodd 1940; Pettey 1948; Zimmermann et al. 2000). Mating occurs one hour before sunrise (Hight et al. 2003) and eggs are laid to form spine-like eggsticks, each with 60-100 eggs. Neonates burrow collectively into cactus cladodes (pads or stems) where larvae feed gregariously and move to new pads as old ones are destroyed. Pupation occurs in plant litter or soil. The moth completes three full generations in Florida, with peak adult flights taking place in April, July, and October (Zimmermann et al. 2004).

Burger (1972) was the first to report on the use of cover sprays of methidathion and carbaryl to protect *Opuntia* plantations in South Africa against attack by both *C. cactorum* and *Dactylopius opuntiae* (Cockerell) (Homoptera: Dactylopiidae). Subsequently, Pretorius et al. (1986) and Pretorius & Van Ark (1992) assayed additional products applied either as cover sprays or stem injections to prevent cladode penetration by first instar *C. cactorum*. Pretorius et al. (1986) indicated that cover sprays of cypermethrin gave excellent results. However, they found that stem injections of monocrotophos gave inadequate control and were expensive and impractical to use against the insect. Pretorius & Van Ark (1992) evaluated additional products (mevinphos and dimethoate) as both stem injections and cover sprays and discovered that these materials applied as sprays translocated effectively through the plants and provided good protection against larval attack. According to Nel et al. (2002), the insecticides currently registered for use against *C. cactorum* in South Africa include a carbamate (carbaryl), an organophosphate (methidathion), and two pyrethroid insecticides (deltamethrin and tralomethrin).

The current infestation of *C. cactorum* in Florida is affecting native *Opuntia* species distributed throughout large expanses of natural areas (*O. stricta* (Haworth) Haworth, *O. humifusa* (Raf.) Raffinesque, and *O. pusilla* (Haworth) Nutall), as well as ornamental cactus plants (*O. ficus-indica* (L.) Miller and *O. stricta*) in urban settings (Hight et al. 2002). Even though chemical control is not a practical or environmentally responsible tactic to protect the millions of hectares of natural *Opuntia* vegetation (Mahr 2001), insecticide controls should be evaluated for their potential use in urban settings. Leibee & Osborne (2001) summarized information on new insecticides to be assayed for use against immature stages of the cactus moth. If proven effective, these products could be employed in culturally managed plantings of *Opuntia* (nurseries, backyards, landscaped public lands) either alone or in combination with other suppression tactics. Furthermore, insecticides could be used to treat ornamental *Opuntia* in nursery settings to ensure that no infested plants are being sold to the public. In this paper we report results of laboratory assays of several insecticides that are registered for use on ornamental plants in Florida. Ovicidal and larvicidal properties of the products were examined and results obtained are discussed in context of the area-wide management of this invasive insect.

## Materials and Methods

### Test Insects

Eggsticks used in these experiments came from a laboratory colony of *C. cactorum* maintained at the USDA-ARS Crop Protection and Management Research Unit, Tifton, Tift Co., GA. The insects are reared on cladodes of *O. stricta* inside rectangular plastic boxes (25 by 17 by 8 cm) that are held in environmental chambers at 26 ± 1°C, a 14:10 (L:D) photoperiod, and 70% RH during larval and pupal development. Cocoons are collected twice per week, de-silked in a dilute bleach solution, and pupae are sorted by gender. Groups of 30-50 newly emerged adults of each gender are placed together in aluminum screen cages (35 by 35 by 35 cm) containing 1-3 cladodes of *O. stricta* for mating and oviposition. Eggsticks are collected from the cages once per day, placed in small plastic cups (60 ml), and maintained at 26 ± 1°C, a 14:10 (L:D), and 70% RH until needed. Under these conditions eggsticks take approximately 30 d to complete their development.

### Products Assayed

Studies were conducted during 2004 at the UF/IFAS North Florida Research and Education Center (NFREC), Quincy, Gadsden Co., FL. Nine different commercially available products were tested in the laboratory for their ovicidal and larvicidal activity against *C. cactorum*. The products were cypermethrin (Ammo® 2.5 E, FMC Corporation, Philadelphia, PA), emamectin benzoate (Proclaim® 5 SG, Syngenta Crop Protection Inc., Greensboro, NC), abamectin (Avid® 1.5 EC, Syngenta Crop Protection, Inc., Greensboro, NC), spinosad (SpinTor® 2 SC, DowAgro Sciences LLC, Indianapolis, IN), azadirachtin (Azatin® EC, AgriDyne Technologies Inc., Salt Lake City, UT), fenoxycarb (Distance® IGR, Valent U.S.A. Corporation, Walnut Creek, CA), imidacloprid (Admire® 2 F, Bayer Corporation Crop Protection, Kansas City, MO), and acephate (Orthene® 75 SP, Valent U.S.A. Corp., Walnut Creek, CA). In addition, the bacterial insecticide *Bacillus thur-*

*ingiensis* Berliner (Dipel®, Valent U.S.A. Corp., Walnut Creek, CA) was evaluated against neonate larvae. Two dilution rates (1.0× and 0.5×) were chosen for each product by averaging the high and low recommended application rates for each material. The average dilution rate was assigned 1.0× and the rate was halved for the 0.5× rate. Only the 1.0× rate was used for *B. thuringiensis*. All products were mixed with de-ionized water and used within 30 minutes of preparation.

### Ovicidal Tests

Cactus moth eggsticks were transported to NFREC where they were divided into sections that contained a minimum of 10 eggs and randomly assigned to treatments. Egg mortality was assessed on both newly laid (1-d-old) as well as on fully embryonated (28-d-old) egg sticks. For each product and dilution rate, eggstick sections were dipped in the treatment solution for 5 s, allowed to air-dry and placed individually inside plastic Petri dishes. Dishes were stored in the laboratory at ambient conditions (25 ± 2°C, 13:11 (L:D), and about 30% RH). Five replicates were completed for each egg age (1-d-old or 28-d-old), insecticidal product (cypermethrin, emamectin benzoate, abamectin, spinosad, azadirachtin, fenoxycarb, imidacloprid, acephate), and dilution rate (1.0× or 0.5×). Controls were dipped in de-ionized water and handled as above. For each experiment, the total number of eggs per eggstick section, the number of eggs that failed to hatch, and the percent mortality was noted per replicate.

### Larvicidal Tests

Only full-size mature eggsticks (28 d old; within 2 d of neonate emergence) were used in these evaluations. Ninety fresh cladodes of *O. stricta* (13 cm in length by 10 cm width) were collected in the field and brought back to the laboratory where the basal joint was allowed to heal before initiating the tests. Ten cladodes were dipped for one min into each product at each dilution rate and allowed to air-dry. Five cladodes of each group were chosen at random and used in the first experiment. The remaining cladodes were stored for 30 d in an outdoor shed at 23 ± 2°C, protected from direct sunlight and rain, and used in the evaluation of residual effects. Decomposition from environmental factors of assayed products on stored cladodes was at a minimum. For both experiments, dipped and air-dried cladodes were placed in plastic containers (14 by 14 by 5.1 cm) with ventilated lids. Individual eggsticks were placed on sections (1 by 2 cm) of filter paper (Whatman #2) on top of each cladode. Containers were held for 14 d in the laboratory under ambient conditions (25 ± 2°C, 13:11 (L:D), and about 30% RH) to allow neonates to emerge and larvae to penetrate the cladode. Results of each experiment were assessed after d 15 by counting the total number of eggs per eggstick and number of eggs that hatched per replicate. Using this information, each cladode was destructively sampled to search for emerged larvae. Five replicates of each product (cypermethrin, emamectin benzoate, abamectin, spinosad, azadirachtin, fenoxycarb, imidacloprid, acephate, and *B. thuringiensis*) and dilution rate (1.0× or 0.5×) were completed for both newly dipped cladodes and cladodes that were dipped and stored for 30 d. Controls were dipped in de-ionized water and handled as above.

### Statistical Analysis

Data from each experiment (ovicidal tests on 1-d-old or 28-d-old eggs and larvicidal tests for newly dipped cladodes and for cladodes that were dipped and stored for 30 d) were analyzed by two-factor analysis of variance (ANOVA) with product and dilution rate as main effects. Interaction between product and dilution rate was included in the model (PROC ANOVA) (SAS Institute 1989). Dependent variables included percent mortality and percent survival, as well as the corrected mean percent mortality with the Schneider-Orelli formula for mortality data from a uniform population (Zar 1984). In addition, arcsine transformed data for each dependent variable were included in the statistical model to satisfy the assumptions of ANOVA. Because no significant effect due to product dilution was detected and because no significant interactions were revealed during the analysis, data for both dilution rates (1.0× or 0.5×) for each product were pooled for each experiment and differences between means were separated by the Waller-Duncan K-ratio *t*-test ($P \leq 0.05$). Likewise, all dependent variables examined yielded similar results and all significant differences in the multiple range tests were the same. Consequently, only data on percent survival of *C. cactorum* in each of the four experiments are presented.

## Results and Discussion

Leibee & Osborne (2001) suggested possible insecticides to screen against the cactus moth. These insecticides are presently registered for use on ornamental plants in Florida and labeled as effective against Lepidoptera that bore into plant tissue (Leibee & Osborne 2001). Six of the nine products suggested by these authors were evaluated in our experiments. The three additional products that we tested were cypermethrin (a synthetic ester pyrethroid) which is extremely effective against *C. cactorum* in South Africa (Pretorius et al. 1986), azadirachtin, a botanical insecticide derived from the neem tree (Meliaceae—*Azadirachta indica* A.

Juss), and the bacterial pesticide *B. thuringiensis* (tested only against neonates).

A summary of our laboratory results is shown in Table 1. Survival of immature stages of *C. cactorum* varied between 64 to 85% when eggsticks were treated with de-ionized water (control). However, one hundred percent mortality (or 0% survival) of 1-d-old eggs was obtained when eggstick sections were treated with cypermethrin, spinosad, or imidacloprid. These products were equally as effective (94 to 100% mortality) when assayed against eggs that were fully embryonated (28 d old), when cladodes of *O. stricta* were exposed to neonates 24 h after dipping, or to cladodes that were dipped and stored for 30 d before exposure. Cypermethrin has been reported to be highly toxic to bees and aquatic insects (US EPA 1989). Pretorius et al. (1986) reported that cypermethrin had good activity against immature *C. cactorum* in South Africa when applied as a cover spray to spineless *Opuntia*. The results of our laboratory assays agree with the data reported by these authors. Spinosad is a macrocyclic lactone insecticide reported to have wide margins of safety for many beneficial insects and related organisms (Schoonover & Larson 1995). Imidacloprid is a nicotinoid insecticide that has minimal environmental and safety concerns associated with its use (Leibee & Osborne 2001). However, it has been found to be acutely toxic to a variety of predatory insects (Mizell & Sconyers 1992).

Emamactin benzoate is an avermectin insecticide that exhibits low toxicity on beneficial insects (Leibee & Osborne 2001). This product was effective at killing eggs and larvae of *C. cactorum* in the laboratory, although some survival of neonates was detected in three of four laboratory assays (Table 1). The second avermectin insecticide that was assayed, abamectin, showed good activity against newly laid and fully embryonated eggs of *C. cactorum*, as well as against neonates that were challenged with newly dipped cladodes. However, the product was ineffective after the cladodes were stored for 30 d. When *B. thuringiensis* was used to prevent neonate penetration into treated cladodes of *O. stricta*, 100% mortality (or 0% survival) was recorded in the laboratory. When we evaluated the results of the assays with *B. thuringiensis*, we found replicates where larvae had been successful at creating an entry hole into the cladode; however, no larvae survived to cause damage beyond this small opening. Finally, azadirachtin, fenoxycarb (a juvenile hormone mimic) and acephate (an organophosphate) were moderately to totally ineffective against immature stages of the cactus moth (Table 1). Lowered effectiveness of some products, such as insect growth regulators (IGRs), may partially be due to feeding behavior of neonate larvae. Eggs hatch synchronously and larvae enter the cladode as a group through a single to few holes. Consequently, few individuals feed on the surface of the cladodes and ingest IGRs sprayed on the surface.

Habeck & Bennett (1990) suggested that widespread use of pesticides was not recommended as a method of control for cactus moth in the Florida Keys because of the occurrence of rare and endangered lepidoptera such as the Schaus swallowtail *Papilio aristodemus ponceanus* Schaus, Florida leaf-wing *Anaea floridalis* Johnson & Comstock and Bartram's scrub-hairstreak *Strymon acis* (Drury). We believe that similar concerns exist for

TABLE 1. EFFECT OF DIFFERENT INSECTICIDES ON PERCENT SURVIVAL OF *CACTOBLASTIS CACTORUM* TREATED AS EGGS THAT WERE NEWLY LAID (1-D-OLD) OR READY TO HATCH (28-D-OLD) AND LARVICIDAL ACTIVITY OF THE PRODUCTS WHEN NEWLY EMERGED NEONATES WERE EXPOSED TO CLADODES OF *OPUNTIA STRICTA* THAT HAD BEEN DIPPED AFTER 24 H OR DIPPED AND STORED FOR 30 D.

| | Mean (± SD) % Survival[1] | | | |
|---|---|---|---|---|
| | Ovicidal Tests | | Larvicidal Tests | |
| Product | Eggs 1-d-old | Eggs 28-d-old | 24 h post cladode treatment | 30-d post cladode treatment |
| Control ($H_2O$) | 80 ± 20.8 a | 85 ± 11.0 a | 64 ± 44.4 a | 81 ± 9.4 a |
| Cypermethrin | 0 c | 0 c | 0 c | 0 c |
| Emamectin Benzoate | 5.8 ± 7.4 c | 0.6 ± 1.9 c | 0 c | 7.9 ± 25.0 c |
| Abamectin | 4.3 ± 9.1 c | 3.3 ± 8.4 c | 0 c | 85.6 ± 8.5 a |
| Spinosad | 0 c | 0 c | 0 c | 0 c |
| Azadirachtin | 52.7 ± 35.1 b | 85.5 ± 11.5 a | 54.6 ± 30.2 ab | 43.6 ± 40.5 b |
| Fenoxycarb | 8.6 ± 27.1 c | 40.3 ± 35.4 b | 64.0 ± 35.2 a | 73.9 ± 13.4 a |
| Imidacloprid | 0 c | 0.6 ± 1.9 c | 0 c | 3.6 ± 10.2 c |
| Acephate | 38.9 ± 33.4 b | 39.7 ± 37.0 b | 35.4 ± 38.7 b | 87.3 ± 10.2 a |
| *B. thuringiensis* | — | — | 0 c | 0 c |

[1]Means within each column followed by the same letter are not significantly different, Waller-Duncan K-ratio *t*-test ($P \leq 0.05$).

all natural areas in Florida and elsewhere in the United States where *Opuntia* are currently infested, or are at risk of being infested, with *C. cactorum*. In these settings, the application of the Sterile Insect Technique (Carpenter et al. 2001; Hight et al. 2004) appears to be the only reasonable management tactic. However, the use of insecticides, together with the removal and destruction of eggsticks, infested cladodes, or entire plants, to protect *Opuntia* in nursery and backyard situations and as a tool to reduce cactus moth pest pressure in urban situations is still recommended. Furthermore, the protection of *Opuntia* plantations destined for fruit or vegetable production in Mexico cannot be overlooked as the insect steadily expands its geographical range to the West.

Our laboratory results suggest possible products that should undergo further evaluations in the field, in particular, *B. thuringiensis*, spinosad, and imidacloprid. However, we would anticipate a much more rapid breakdown in the effectiveness of *B. thuringiensis* in the environment due to increased exposure to UV light and rain events. Because these products are already registered for use on vegetables and ornamental plants in Florida, expanding their registration in other states is highly recommended and could perhaps lead to the eventual acceptance of these products for use in fruit and vegetable plantations of *Opuntia* in Mexico. Lastly, when formulations become available, field tests are recommended for isolates of AcMNPV, a nuclear polyhedrosis virus isolated from *Autographa californica* (Speyer) (Lepidoptera: Noctuidae). This islolate has been shown by Vail et al. (1984) to be moderately effective against immature stages of *C. cactorum* in the laboratory.

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