

**CHEMICAL CONTROL OF
TARNISHED PLANT BUG -
RESULTS FROM FIELD CAGE STUDIES
AND LABORATORY BIOASSAYS**

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Abstract

Laboratory bioassays were conducted to evaluate response of tarnished plant bug to insecticides that induce changes in feeding behavior. Both imidacloprid and pymetrozine administered to adults in a sucrose/dye mixture resulted in reductions in excretion of fecal deposits over 4 and 8 day test periods. A conventional poisoning effect was observed at higher concentrations for imidacloprid. Results from field trials with caged tarnished plant bugs indicate additions of sucrose or molasses to spray mixtures of Provado improve efficacy at low rates (0.02 to 0.025 lb a.i./ac) but not at higher rates (0.045 lbs a.i./ac). Data suggest that conventional spray and count methods are not sufficient to adequately evaluate insecticides with novel modes of action that affect feeding behavior.

Introduction

Tarnished plant bug, *Lygus lineolaris* (Palisot de Beauvois), is a key pest of cotton in the Midsouth states of Arkansas, Louisiana and Mississippi. The insect can cause damage to the plant terminal and squares resulting in delayed crop maturity (Tugwell et al. 1976, Hanney et al. 1977, Smith 1985). Chemical control remains the standard method for managing tarnished plant bug, but populations resistant to the major classes of insecticides have been identified in the Midsouth (Snodgrass and Scott 1988; Snodgrass 1994; Snodgrass and Elzen 1995). In this production region, continued efficient crop protection from plant bug injury will require pest management programs that make wise use of available tools and, as they become available, incorporation of new classes of efficacious insecticides.

We report here on field and laboratory trials with tarnished plant bug conducted in 1995. A significant research effort was focused on developing a simple, low cost technique for evaluating insecticides with oral activity against tarnished plant bug and determining their effect on feeding behavior. Laboratory work included studies with the chlornicotinyl, imidacloprid (Provado, Bayer, Inc., Kansas City, KS) and the pyridine azomethrine, pymetrozine (CGA 215944,

Ciba Crop Protection, Greensboro, NC). Imidacloprid has high oral activity against many sucking and chewing insects (Elbert et al 1991). It has been shown to produce conventional poisoning effects as well as antifeeding and repellency effects against aphids (Nauen 1995). Pymetrozine is not directly toxic to sucking insects such as aphids, but it has been shown to affect the nervous system that controls feeding behavior (Fluckiger et al. 1992; Denholm et al. 1995). Our field work included evaluations to determine if additions of "sweeteners" would affect efficacy of imidacloprid.

Materials and Methods

Laboratory Trials

All laboratory evaluations were performed with adult tarnished plant bugs collected in Craighead Co, AR on wild hosts in areas where no insecticide applications had been made. During testing, insects were held at 80 ° F and 70 to 85 % RH.

Bioassays were made using a single adult in a 100 X 4 mm plastic petri dish with the test material mixed with 10% sucrose solution containing either a blue or green marker (McCormick food coloring - 6 drops/50 ml). The toxin/sucrose/dye mixture was placed in the petri dish either in bubbles made from Parafilm M™ (American National Can™, Nenah, WI) or on cubes (1 cm³) cut from the green foam developed for the floral industry to provide water to cut flowers (Water Foam from Styrofab, Waxahatchie, TX). For the bubble procedure, a 96-well cell culture plate (35mm cell diameter) was used as a form for shaping the bubbles. A 4 X 6 inch strip of Parafilm was laid over the plate, and round end map pins with heads size slightly smaller than the well diameter were used to stretch the parafilm down into the well. Approximately 70 microliters of the test mixture was pipetted into the newly made "bubble". Another sheet of Parafilm was stretched over the bottom sheet and a round, rolling pie crust shaper was rolled over the 2 sheets to seal the edges around each well. Individual bubbles were cut out with scissors and 1 was placed in each petri dish.

Fecal deposits from adults feeding on the dyed test materials are easily observed in the petri dish. In these trials fecal specks ("freckles") were counted daily. At the end of specified test periods, usually 3 to 4 days, freck production per individual was categorized as zero, low (1 or 2), medium (3 to 4) and high (>4).

Feeding trials with imidacloprid (75 WP) were initiated on 22 June with mixed aged wild adults that had been held 24 hrs with broccoli and 10% sucrose as food. Parafilm bubbles were prepared with imidacloprid at concentrations of 0, 0.1, 0.5, 1, 5 and 10 ppm (a.i.) mixed with 10 % sucrose to which green food coloring had been added. In addition to the bubble, a water moistened 47 mm diameter pad (Gelman pad, product # 66025, Gelman Sciences, Ann

Arbor, MI) was added to provide water. There were 15 insects tested at each concentration. Mortality was determined at 24, 72 and 96 hrs.

Additional feeding trials using parafilm bubbles were initiated 3 July with 3 day old adults that had been reared from field collected nymphs reared to adult stage in the laboratory on broccoli. Parafilm bubbles were prepared as above with imidacloprid at concentrations of 0, 0.01, 0.05, 0.1 and 0.5 ppm (a.i.). A water moistened 6 mm diameter Gelman pad was added to provide water to the insect. There were 25 to 30 insects tested at each concentration. Freck counts were made at 72 hrs. Mortality was determined at 72 and 96 hrs.

A pymetrozine and imidacloprid bioassay using water foam cubes was initiated 18 Aug. Treatments were pymetrozine (50% WP) at 0, 0.1, 1, 10, 100 and 1000 ppm (a.i.) and imidacloprid (75%) at 0.5 ppm (a.i.) prepared with 10% sucrose mixture + green food coloring. Ca. 1.25 ml of the appropriate insecticide/dye mixture was pipetted on a water foam cube and placed with one adult tarnished plant bug. Test insects were adults collected on wild hosts and held 24 hrs on 10 % sucrose (no dye) moistened paper towels at 60° F. At 4 days all cubes were replaced with cubes containing 10% sucrose and blue food coloring but no insecticide. With this method, freck production in the first 4 days with toxin (green frecks) could be differentiated from feeding on sucrose alone in days 5 through 8 (blue frecks). In all trials, insects were scored dead when no movement was detected in 5 seconds after the insect had been probed with a camel's hair brush.

Field Trials

Studies were conducted with imidacloprid in combination with "sweeteners", sucrose and feed grade molasses, at the UA Cotton Branch Experiment Station in Marianna, AR and the ASU Research Farm in Jonesboro, AR. Numbers of native tarnished plant bug were too low in 1995 to conduct whole plot evaluations so cage tests were initiated.

For each trial, 3 organdy sleeve cages, 9 inches diam by 22 inches long, were secured to individual plants in each plot by tying the lower end of each cage around the plant ca 1 ft from the terminal with twist ties. The cages were rolled down to the tie and covered with aluminum foil. Following application the foil was removed, the cage pulled up, and 5 field collected insects were placed into each cage.

Cage tops were secured with twist ties. Insects had been collected by sweep net on mustard immediately prior to testing and held in 15 ml plastic vials (5/vial) on ice. After 72 hrs plants were cut below the cage and taken to the laboratory where mortality was determined.

The field study in Marianna was conducted to determine effect of sucrose as a spray additive on efficacy of imidacloprid (Provado). The variety DPL 51 was planted on 15 May 1995 into plots 6 rows (38 inch centers) wide

and 50 ft long separated by 2 row buffer zones. Treatments were Provado 1.6 F (0.025 and 0.047 lbs a.i./ac), Provado 1.6 F (0.025 and 0.047 lbs a.i./ac) tank mixed with 10 % sucrose solution (granulated pure cane sugar), Orthene 90 S (0.5 lbs a.i./ac) (acephate, Valent USA Corporation, Walnut Creek, CA) and an untreated control. Treatment plots were arranged in a RCBD with 3 replications. Insecticides were applied 21 July using a 4-row CO₂ charged back pack sprayer calibrated to deliver 13 gpa at 23 psi with 1 TJ-50 8002vs nozzle per row. Test insects were collected on blooming mustard adjacent to the field.

In the Jonesboro trial, effects of additions of feed-grade molasses to Provado were evaluated. Cotton (DPL 51) was planted 4 June 1995 on the ASU farm in 4 row (38 inch centers) wide plots 60 ft long separated by a 4 row buffer. Treatments were Provado 1.6 F (0.022 and 0.047 lbs a.i./ac), Provado 1.6 F (0.022 and 0.047 lbs a.i./ac) tank mixed with molasses (2 qts/25 gal water), Orthene 90 S (0.5 lbs a.i./ac), molasses alone and an untreated control. Molasses was obtained from a local cattle feed mill. Treatments were arranged in a RCBD with 3 replications. Insecticides were applied 8 Aug using a 4-row CO₂ charged tractor mounted sprayer calibrated to deliver 12.5 gpa at 40 psi with TJ-50 8002 nozzles on 17 inch spacing. Test insects were collected adjacent to the field in blooming mustard.

Results

Laboratory Bioassays

In conventional insecticide bioassays with OPs and synthetic pyrethroids with tarnished plant bug, mortality readings are taken at 24 and 48hrs (Snodgrass and Elzen 1995). Extending the holding period to 96 hrs was necessary in our evaluations with imidacloprid to allow us to examine both conventional poisoning effects and possible sublethal effects of the compound.

Tarnished plant bug mortality data from the 22 June trial indicated a dosage-response relationship at 24 hours with imidacloprid at rates above 1ppm (Figure 1). These results suggest a conventional poisoning effect at these high concentrations. At concentrations less than 1 ppm, mortality was very low in the 24hr readings but increased substantially by 96hrs.

Results from the 3 July trial indicate that response to imidacloprid included a reduction in freck production in the first 3 days of the feeding at the 0.5ppm concentration (Figure 2). We interpreted reduced freck production to be associated with a reduction in feeding activity.

Mortality data from the 18 Aug evaluation indicate no evidence of a dose- mortality relationship with pymetrozine after 4 days. With an extended holding period (8 days) mortality levels rose to 88% at the 1000ppm concentration (Figure 3). Although survival rates were above 50% for all

treatments at 4 days, reductions in freck production were observed in the imidacloprid treatment and with pymetrozine concentrations at 1 ppm and above (Figure 4). By 8 days, measurements of freck production indicate that when imidacloprid treated insects were provided diet containing no toxin in days 4 through 8, feeding resumed for the surviving insects. Data are unclear and ambiguous as to whether there was recovery among pymetrozine treated insects.

Field Trials

Addition of sugar to the low rate of Provado significantly increased mortality over 3 days compared to the same rate without sugar (Figure 6). Similar results were observed with additions of molasses to low rates of Provado (Figure 7). At the 0.047 lb/ac rate, no differences in mortality levels were observed with addition of sugar or molasses. Highest mortality (100%) was observed with Orthene in both trials.

Discussion

Nauen (1995) reported an antifeedant effect with sublethal concentrations of imidacloprid in cotton with green peach aphid (*Myzus persicae* (Sulz.)) which resulted in reduction of honeydew excretion. Our results from tarnished plant bug studies show that freck production is reduced in individuals fed concentrations of imidacloprid that do not immediately result in mortality (sublethal doses). This effect appears to be reversible because individuals will begin to feed again following exposure if an alternative food source are available. It is unknown if the reduction in feeding is due to repellency or intoxication; both have been implicated in feeding studies with aphids (Nauen and Elbert 1994).

In work with pymetrozine, Denholm et al. (1995) recommended extending the duration of bioassays with cotton whitefly (*Bemesia tabaci*) and cotton aphid (*Aphis gossypii* Glover) to 120hr following initial exposure. A dosage response with cotton whitefly was apparent if evaluation times were extended to 120h and mortality reached 100% at 100 ppm, but they reported little direct mortality with cotton aphid adults. Because pymetrozine acts primarily through prevention of feeding, they stated that the time required to evaluate cotton aphid adult mortality appeared to be determined solely by the period for which exposed individuals could survive without further food. Possibly this also could be the case with tarnished plant bug, but further study is required to determine this. Our results indicate that tarnished plant bug do not appear to be as sensitive to pymetrozine as cotton whitefly, but at rates above 1 ppm, some reduction in feeding did occur following 4 days exposure.

In field cage bioassays tarnished plant bug mortality from imidacloprid was increased at low rates if a "sweetener" was included in the tank mix. No increase was observed at

higher rates. This may be an indication of a repellency effect at the higher rate.

Evaluation and user acceptance of new insecticides is simplified if the products have conventional poisoning effects that are easily observed in the field using standard spray and count evaluations. For insecticides with novel modes of action, conventional evaluation methods may be insufficient. This is especially true if crop protection benefits are achieved partially or wholly through pest repellency or through antifeeding effects. With these products live insects can remain in the field after application, even though they may no longer damage the crop. Evaluation criteria with these type of products will require adoption of an expanded array of crop protection measures to augment traditional spray and count methods. For tarnished plant bug in cotton, this could include monitoring plant damage and square shed.

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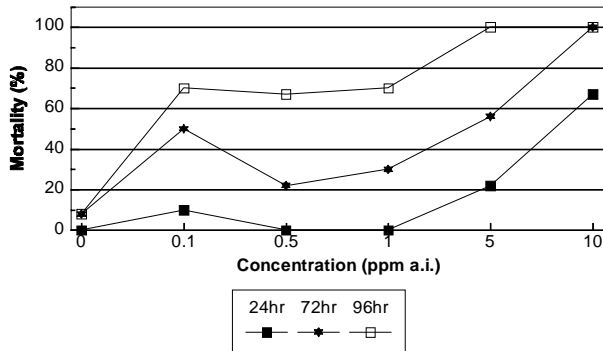


Figure 1. Mortality of tarnished plant bug adults 24, 72 and 96 hrs after feeding with imidacloprid/sucrose/dye mixtures presented in parafilm bubbles.

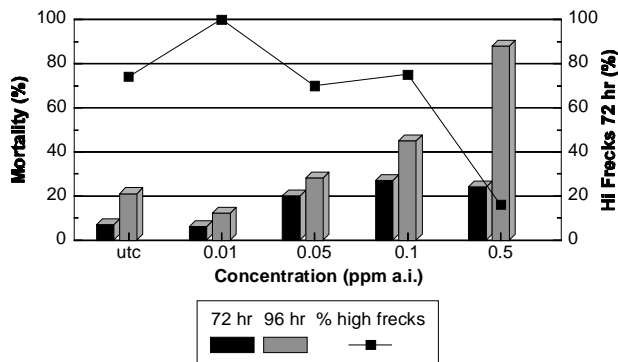


Figure 2. Mortality of tarnished plant bug adults at 72 and 96 hrs and % insects with high freck counts at 72 hrs after feeding with imidacloprid / sucrose / dye mixture presented in parafilm bubbles.

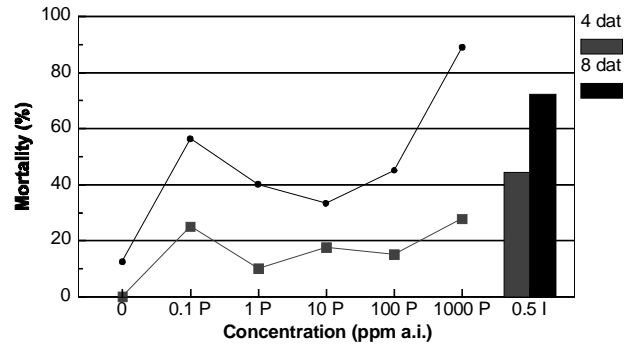


Figure 3. Mortality of tarnished plant bug adults after feeding on pymetrozine (P) or imidacloprid (I) presented in a sucrose/green dye mixture on water foam cubes which were replaced after 4 days with cubes containing sucrose/ blue dye mixtures (no toxin).

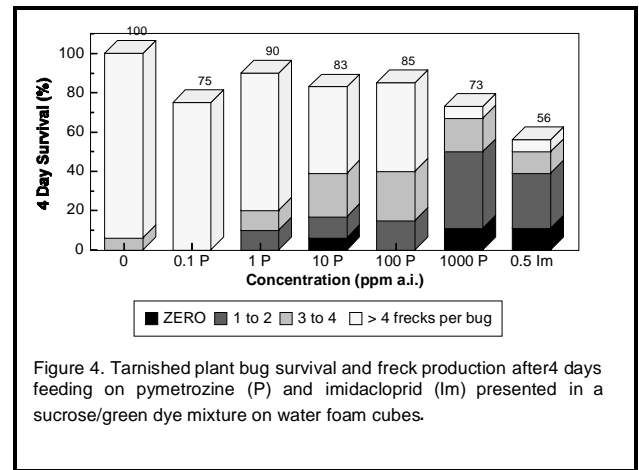


Figure 4. Tarnished plant bug survival and freck production after 4 days feeding on pymetrozine (P) and imidacloprid (Im) presented in a sucrose/green dye mixture on water foam cubes.

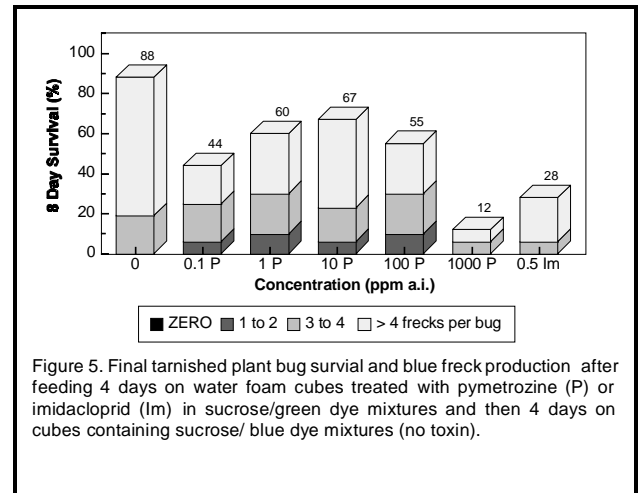


Figure 5. Final tarnished plant bug survival and blue freck production after feeding 4 days on water foam cubes treated with pymetrozine (P) or imidacloprid (Im) in sucrose/green dye mixtures and then 4 days on cubes containing sucrose/ blue dye mixtures (no toxin).

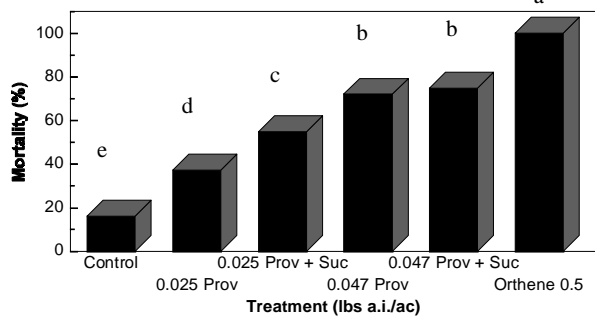


Figure 6. Effect of sugar additive in spray mixtures with imidacloprid (Provado) on mortality of caged tarnished plant bugs in field plots. Bars carrying the same letter are not significantly different ($P>0.05$: least significant differences).

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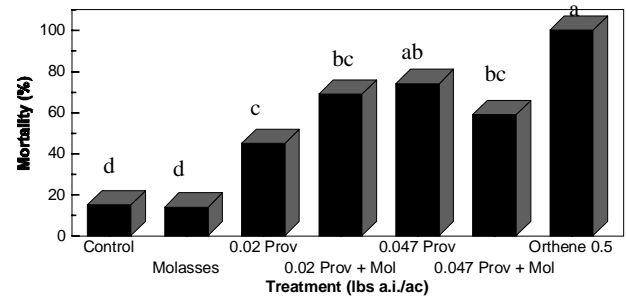


Figure 7. Effect of molasses additive in spray mixtures with imidacloprid (Provado) on mortality of caged tarnished plant bugs in field plots. Bars carrying the same letter are not significantly different ($P>0.05$: least significant differences).