Colegio de Postgraduados

Moth trapping of males using *Diatraea* spp. (Lepidoptera: Crambidae) pupae in sugarcane plantations

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ABSTRACT

Objective: To standardize the moth trapping method through the use of Delta traps with *Diatraea* pupae to lure males.

Design/Methodology/Approach: The following lure types were placed inside Delta traps: two female pupae (T1), three female pupae (T2), and two female pupae with one adult female (T3). The response variable was the number of male moths captured per trap. An analysis of variance and Tukey's test ($p \leq 0.05$) were performed.

Results: Statistical differences ($p \leq 0.05$) were observed between T3 and T1/T2: T3 trapped the lowest number of specimens (0.94 adults/trap) from May to July 2022. Likewise, June recorded the highest abundance of *Diatraea* moths (3.05 adults/trap). Nevertheless, T1 captured the highest number of specimens (4.16).

Study Limitations/Implications: The use of commercial pheromones has been established as a useful moth trapping method in the case of stem borers. In recent years, no favorable results have been observed with the use of pheromones of the *D. saccharalis*, *D. grandiosella*, and *D. considerata* species in Delta traps. These findings could be indicative of the presence of another *Diatraea* species.

Findings/Conclusions: The use of two *Diatraea* pupae per trap attracts male moths. This method would help to detect and regulate moths, with the aim of interrupting their life cycle.

Keywords: Sex pheromones, pest, stem borer.

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INTRODUCTION

The stem borer is widely spread across the American continent and is a major pest of corn (*Zea mays* L.) and sugarcane (*Saccharum* spp.) (Lastra and Gómez, 2006). Various species of stem borers (*Diatraea*, *Eoreuma loftini*, and *Elasmopalpus lignosellus*) have been reported in Mexico; in their larva stage, they cause yield losses of 2 to 10 tons of sugarcane per hectare (Rodríguez and Vejar, 2008). Different species of genus *Diatraea* have been reported, increasing the complexity of their management. The damage they cause can be direct and is reflected in dead hairs or "pelillos" (primary shoots), stems, and bored buds, potentially reducing field yields by up to 50% (CONADESUCA, 2021).

Meanwhile, the damage caused by borer larvae to juice quality can be worsened by the proliferation of fungi and bacteria, which enter the plant through the cavities made by the larvae from within the stem. Among the fungi reported are *Colletotrichum falcatum*, *Fusarium*, and *Nigrospora* (Joyce *et al.*, 2016).

The use of synthetic pheromones has successfully managed stem borer populations. Nevertheless, a low number or no specimens of the *Diatraea* species (*D. saccharalis*, *D.*

grandiosella, *D. considerata*) have been trapped in recent years. In contrast, the use of live insects in pupal and adult stages has successfully determined the minimum number of individuals required to trap male moths for each study region (Assis *et al.*, 2022). The objective was to determine the number of pupae per trap required to trap *Diatraea* male moths in sugarcane plantations.

MATERIALS AND METHODS

Study area. The study was conducted in the *ejido* Palmillas, municipality of Yanga, Veracruz, Mexico, part of the sugarcane supply area of the Central El Potrero sugar mill (18° 49' 08.9" N and 96° 46' 38.8" W).

Biological material. Larvae of the genus *Diatraea* were collected in the Palmillas study area, before the start of the experiment. They were subsequently transferred to the laboratory of Applied Microbial Biotechnology at the Colegio de Postgraduados - Campus Córdoba. A rearing of *Diatraea* larvae was established inside the facilities, under aseptic and controlled conditions, with a 12:12 (light:dark) photoperiod at 28±2 °C (Lastra and Gómez, 2006). Each larva was placed in a separate 500-mL transparent plastic container along with a 10-cm long cane stalk. The larvae were kept under rearing conditions, until they reached the pupae stage. The specimens were then sexed (Butt, 1962) and selected for reproduction (female and male). Only the female pupae were used in the field attraction evaluation (Figure 1).

Treatment installation

The experiment was set up in the first week of May 2022, when the crop was six months old (ratoon cycle 2: R2). The 5-ha productive area was divided into three plots, according to the proposed experimental design. The following lures were placed within the traps: two female pupae (T1), three female pupae (T2), and two pupae and one female adult (T3), as the dependent variable. The traps were placed 26 m apart from each other and the distance between repetitions measured approximately 60 m.

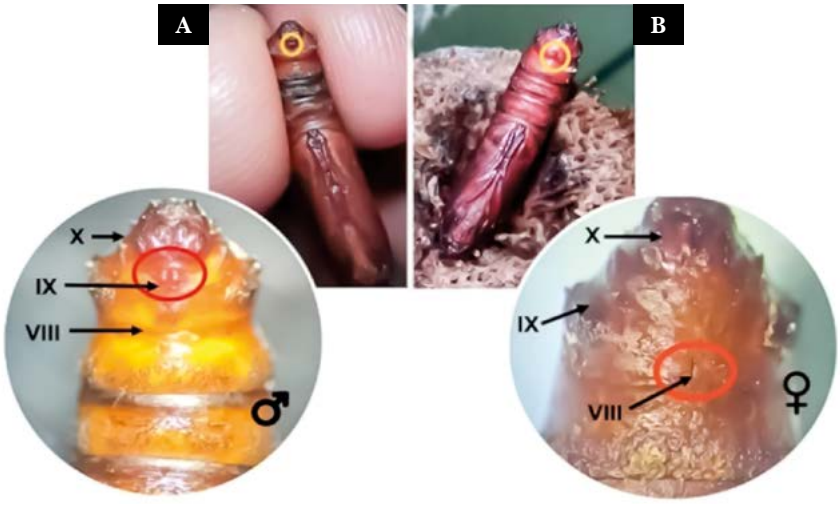

Figure 1. Sexed pupae of *Diatraea*: A) male pupa and B) female pupa.

A trap was installed for each treatment, which consisted of a Delta-type trap (29 cm deep × 21 cm wide × 17 cm high) with a sheet impregnated with an Adhequim® 100 organic polymer. A Reyma® 4-ounce transparent plastic container, covered with anti-aphid mesh, was placed in the center of the trap, where the live insects were deposited (Assis *et al.*, 2022).

Determination of response variables

The moths caught in the Delta traps were monitored weekly. The polymer sheet was replaced every 15 days. Captured male moths were recovered from the trapping film with a paintbrush and white gasoline. Male moths were placed in 50-mL Corning® plastic centrifuge tubes and kept refrigerated at −20 °C for later identification.

Experimental design and data analysis

A randomized complete block design with three replications was established. The treatment consisted of a two-factor factorial design: 1) the effect of the traps with different female pupae and 2) the time (months) during which the measurements were taken.

The response variable was the number of male moths captured per trap. The captures were recorded every 15 days and monthly averages were calculated. The resulting data were subjected to the Shapiro-Wilk normality test and Bartlett's test for homogeneity ($p \leq 0.05$). To determine the treatment effect, an analysis of variance (ANOVA) under the proposed design and Tukey's test ($p \leq 0.05$) were performed in Minitab v. 21 for Windows.

RESULTS AND DISCUSSION

The ejido Palmillas —where the experiment was conducted— had a history of high percentages of damage (>5%) caused by *Diatraea* (Cruz-Tobón *et al.*, 2023). Prior to the experiment, traps with synthetic hormones were set up to lure *D. saccharalis*, *D. grandiosella*, and *D. considerata*. Traps with live insects (natural pheromone) were also prepared. Traps with synthetic pheromone did not capture moths of any of the above-mentioned species, whereas the trap with live insects successfully captured male moths (Assis *et al.*, 2022).

No statistical differences were detected between T1 and T2 ($p \leq 0.05$), with average captures of 3.05 and 2.72 adults/trap, respectively. Meanwhile, T3 had significant differences with respect to the rest of the treatments, with 0.94 adults/trap captured in the treatment with two pupae and one adult. In conclusion, the addition of a female adult moth has an adverse effect, reducing moth trapping by 300%. Consequently, T1 with two female pupae could be the best treatment for subsequent evaluations with pupae (Figure 2).

Previous studies have reported successful results using Delta traps with three pupae and three newly emerged female moths (Butt, 1962) as luring strategies for the management of *Diatraea* spp., populations.

Assis *et al.* (2022) and other authors have recommended setting traps with two female moths 500 m apart from each other. The adults trapped with this device will help to initiate the release of parasitoids.

Species of genus *Diatraea* account for high percentages of the damage caused to sugarcane plantations. Therefore, interrupting the life cycle of male moths and limiting the frequency of male-female copulation is an alternative for the regulation of the stem

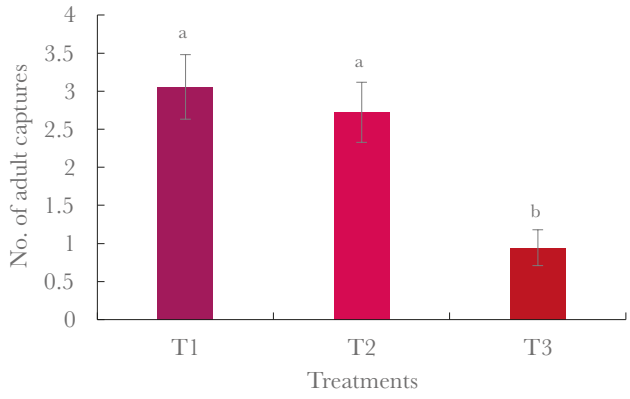

Figure 2. Number of adults per treatment. The bars show the mean ± standard error. Means with different letters are significantly different (Tukey, $p \leq 0.05$).

borer larvae of a given species, reducing the oviposition of viable eggs. As a consequence of the trapping of male moths, less larvae will emerge. Subsequently, the positive relationship between the presence of trapped moths and the percentage of damage caused by larvae detected per hectare could be estimated. Wilson *et al*. (2012) used the same method to control *E. loftini*.

Regarding the time effect, significant differences were observed in July, when less *Diatraea* adults were recorded (one adult per trap) than in May and June, when three individuals were recorded per trap (Figure 3).

The heat map shows the relationship between the treatment and time variables, using color gradient (number of trapped specimens) (Wilkinson and Friendly, 2009; Akers, 2015). In conclusion, the treatment with two female pupae (T1) had the highest average number of captures and the highest presence of male moths occurred in June.

In other words, the increased abundance of adult *Diatraea* populations is related to two main factors: 1) the age of the crop and 2) the developmental stage of the insect. In the first case, the presence and abundance of *Diatraea* is associated with young crops with succulent

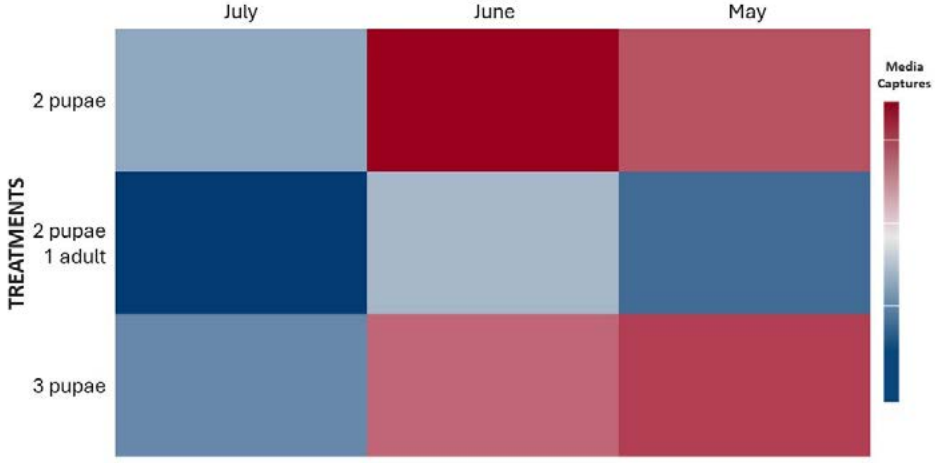

Figure 3. Heat map (Minitab version 21).

and fibrous stems which allow their first-instar larvae to bore and penetrate the plant (Cruz-Tobón *et al.*, 2023). In the second case, *Diatraea* need in average 42-60 days to complete their cycle, according to Gómez and Vargas (2014). Therefore, the damage produced by the larvae must have occurred from April to May and adults were observed in June, which matches the findings of this study. Damage can also be the result of the agronomic management of the crop or environmental factors, such as humidity, precipitation and temperature (Pannuit *et al.*, 2015; Carbognin *et al.*, 2023).

CONCLUSIONS

Traps with two pupae enable the capture of male moths. In addition, this strategy can determine the presence of *Diatraea* species and manage their populations by facilitating an efficient and specific trapping. The timing and distribution of traps are fundamental for the regulation of *Diatraea* populations and are an efficient management strategy. In conclusion, this study determines the possibility of further evaluations about the reach of pheromones as a live lure and specifies the distance and number of traps per hectare.

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