

Sexual pheromone for monitoring *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in cotton crops in Mato Grosso, Brazil

By

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Abstract

Cotton crop, despite its great relevance in Brazil, especially in Mato Grosso, lacks more effective technologies for monitoring and controlling *Spodoptera frugiperda*, one of the most widespread pests currently affecting the crop. In this context, pheromones represent a promising tool for the management and monitoring of this pest. Therefore, this study aimed to evaluate the comparative efficiency of different synthetic sex pheromone formulations for monitoring *S. frugiperda* in cotton crops. The research was conducted in a cotton field located in Tangará da Serra – MT, during a ten-week period. Delta-type sticky traps were used in a randomised experimental design with five treatments and five replications: 3 experimental formulations, virgin females (positive control) and hexane (negative control). The variables analysed included the total number of adults captured per trap and the proportion of males and females. Data were subjected to analysis of variance, and treatment means were compared using a significance test at 5% probability. During the ten weeks of evaluation, 3,660 moths of *S. frugiperda* were captured, of which 3,548 were males (96.94%) and 112 were females (3.06%). Capture by the formulations did not show a significant difference; however, they differed from the positive control (virgin females). The results indicate that the synthetic formulations tested present similar efficiency for monitoring *S. frugiperda* in cotton crops. However, more detailed studies with strains from Mato Grosso are necessary for the isolation and identification of pheromonal compounds from local populations, aiming to improve the effectiveness of monitoring strategies for this pest.

Keywords: Fall Armyworm, Field trapping, Integrated Pest Management, Regionalization of Pheromones.

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Introduction

Brazil stands out in the global scenario of cotton *Gossypium hirsutum* Linnaeus, 1753 (Malvaceae) production and is currently among the five largest producers in the world (USDA, 2025). In 2025, Brazilian cotton production was estimated to occupy an area of 2,137,100 ha, with an average productivity of 1,885 kg ha⁻¹ and a total production of 4,027,900 t, representing an increase of 2.4% in cultivated area, despite a reduction of 3.6% in productivity and 1.2% in total production compared to the previous season (CONAB, 2025). For the state of Mato Grosso, Brazil's main producing region, the cultivated area in 2025 was estimated at 1,434,825

ha, with a total production of approximately 6,257,347 t (IMEA, 2025).

Despite the high productivity of cotton crops, phytosanitary problems caused by a complex of more than 30 arthropod species can lead to significant yield losses and increased production costs, mainly due to the intensive use of broad-spectrum insecticides (Rolim & Netto, 2021). According to the Instituto Mato-Grossense de Economia Agropecuária (IMEA, 2025), in the 2023/24 cotton season costs amounted to US\$ 4.30 thousand per hectare, in 2024/25 the cost was US\$ 4.01 thousand/ha, and in the 2025/26 season it increased to US\$ 4.67 thousand/ha.

Among insect pests, the caterpillar *Spodoptera frugiperda* (J.E Smith, 1797) (Lepidoptera: Noctuidae) stands out for the damage it initially causes to the leaves, reducing the photosynthetic area and later destroying flower buds and developing bolls (Barros et al., 2010). It is a polyphagous species that presents high mobility, fecundity, and pupae with facultative diapause (Nagoshi, 2009). 76 families of host plants are described, including species of economic interest such as soybean, corn, and cotton (Montezano et al., 2018). The succession of these cultures, the ease of migration and survival favor proliferation and make it difficult to control (Montezano et al., 2018).

Chemical control of agricultural pests has been widely based on the intensive use of insecticides, often applied in a non-selective manner. This practice negatively affects non-target organisms, including pollinators and natural enemies, which play an essential role in maintaining ecological balance in agroecosystems (Lopes & Albuquerque, 2018).

Moreover, the excessive and recurrent use of chemical insecticides has favored the selection of resistant populations. In Brazil, cases of resistance in insect pests have already been reported for active ingredients such as lambda-cyhalothrin (pyrethroid), chlorpyrifos (organophosphate), chlorantraniliprole (diamide) and lufenuron (benzoylurea), according to the Insecticide Resistance Action Committee (2023).

Sustainable pest management practices have increasingly focused on alternatives to conventional chemical control, including the use of semiochemicals such as sex pheromones. These compounds are produced by insects to attract individuals of the opposite sex of the same species and can be applied as an effective tool for pest monitoring in agricultural systems (Araújo et al., 2021).

Pheromone-based monitoring stands out as a highly specific method, as it involves the release of volatile molecules in the field to attract and capture target moths. The number of individuals captured in traps provides valuable information on population density, supporting more accurate and timely decision-making for pest management (Muthukumar & Kennedy, 2021).

The pheromonal compounds of *S. frugiperda* responsible for the attractiveness of males are the acetate esters of (Z)-9-tetradecenyl acetate (Z9-14:Ac), (Z)-11-hexadecenyl acetate (Z11-16:Ac), (Z)-7-dodecenyl acetate (Z7-12:Ac), (Z)-9-dodecenyl acetate (Z9-12:Ac) and (E)-7-dodecenyl acetate (E7-12:Ac), with Z9-14:Ac reported as the major component (Batista-Pereira et al., 2006; Cruz-Esteban, 2020), and some of these compounds are already part of commercial formulations (Bratovich et al., 2019; Sharath et al., 2022).

Pheromonal compounds from a given population, even when presenting the same chemical composition, do not necessarily show the same efficiency in populations from different geographic regions (Andrade et al., 2000; Cruz-Esteban et al., 2018; Unbehend et al., 2014), which highlights the importance of testing pheromone-based formulations under

local production conditions. Thus, the objective of this study was to evaluate the comparative field efficiency of different synthetic sex pheromone formulations for capturing and monitoring populations of *Spodoptera frugiperda* in cotton crops, under the edaphoclimatic conditions of the municipality of Tangará da Serra, Mato Grosso.

Materials and methods

Study area

The experiment was conducted at Colorado Farm, located in Tangará da Serra, Mato Grosso, Brazil (14°38'01.8"S, 57°38'14.0"W), during the early reproductive stage of the 2019/20 cotton season. The study was carried out in a 177.7 ha field planted with the Bt cultivar FM 954 GLT, while the border rows were sown with the conventional cultivar FM 944 GL as a refuge area. Pheromone traps were installed along the field margins, exclusively within the conventional cotton. The region has an average annual rainfall of 1,830 mm and a mean air temperature of 26.1 °C (Daniel et al., 2021; Dallacort et al., 2011). The formulations were supplied by the Laboratory of Natural Products Chemistry Research (LPQPN) at the Federal University of Alagoas (UFAL).

Obtaining the formulations

Based on information available in the Pherobase for *S. frugiperda*, three synthetic pheromone formulations were produced at the Laboratory for Research in Natural Resources of the Federal University of Alagoas. Two of these formulations shared the same chemical composition but differed in the total amount of pheromone incorporated, allowing an evaluation of the influence of dose on moth attraction. The third formulation also maintained the main pheromonal components but included the addition of a plant-derived volatile compound with known food-attractant properties, aiming to enhance the attractiveness of the blend. In all treatments, an antioxidant was incorporated to ensure chemical stability and prevent compound degradation during field exposure (Table 1).

Table 1. Synthetic formulations prepared for capturing *Spodoptera frugiperda* moths (Lepidoptera: Noctuidae) in cotton crop in Mato Grosso, Brazil, in March of 2020.

Formulations	Components (%)				
	(Z)-9-tetradecenyl acetate	(Z)-11-hexadecenyl acetate	(Z)-7-dodecenyl acetate	(E)-2-hexenal	α-tocopherol
1 (3 mg)	72.3	18.00	4.70	-	5.00
2 (6 mg)	72.3	18.00	4.70	-	5.00
3 (6 mg)	70.3	18.00	4.70	2.00	5.00

Insects rearing

To obtain virgin females, a laboratory colony was established at the Entomology Laboratory of the State University of Mato Grosso (UNEMAT), using caterpillars collected from a cotton field. Larvae were reared under controlled conditions on a semisynthetic diet adapted from Greene, Leppla & Dickerson

(1976), which provides a balanced mixture of proteins, carbohydrates, vitamins, minerals and a solidifying medium, a nutritional composition designed to support normal development comparable to natural foliar diets. Emerging females from the first laboratory generation were used as the control treatment.

Field experiment

The experiment was carried out during the cotton harvest (2019/2020). The border area of a plot with the conventional cultivar FM 944 GL in the reproductive stage was used. It should be noted that the area was bordered by conventional and Bt corn (Figure 1).



Figure 1. Aerial image of the experiment implementation site (A) and Trap containing septum with pheromone (B) to attract males of *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in cotton crop of Mato Grosso in the 2020/2021 harvest.

For field collection, Delta-type sticky traps, white in color, were used. The formulations were incorporated into rubber septa inserted in the center of the sticker (Figure 1B). The traps were distributed along the border, installed on stakes above canopy according to plant growth. The traps (19x18x20cm) were obtained from the company Isca Tecnologias Ltda.

The experimental design was completely randomized, with five treatments and five replications: three synthetic formulations described in the study (Table 1); five virgin females (Blassoli-Moraes et al., 2016), aged 0–48 h and kept in cages as a positive control; and hexane P.A. as a negative control.

In total, 25 traps were distributed in the field in a refuge area bordered by Bt cotton and corn crops. The traps remained in the field for ten weeks, being installed on May 4, 2020. The septa were replaced on June 18, 2020. Evaluations were carried out weekly. Virgin females and sticky cards were replaced, and the traps were relocated to avoid positional bias. Cotton plants were randomly inspected to detect the presence of caterpillars in the border area (Figure 1). Captured moths were identified using illustrated keys (Michereff Filho et al., 2019). It is important to note that the farm maintained its pesticide application protocol throughout the evaluation period.

Data analysis

The effects of experimental treatments on male capture were analyzed using a General Linear Model with a Poisson

distribution, implemented via the GLM function. Subsequently, a Tukey test with a 5 % significance level was conducted using the General Linear Hypotheses (glht) function from the multicomp package (Hothorn et al., 2008). To enhance the understanding of capture patterns, analyses of weekly averages for each treatment were performed. Concurrently, a control level of three moths per trap per night, as defined by Cruz et al. (2012), was considered across evaluation intervals. All analyses were executed in R version 3.5.1 (R Core Team, 2023), and graphical representations were created using the ggplot2 package (Wickham, 2016).

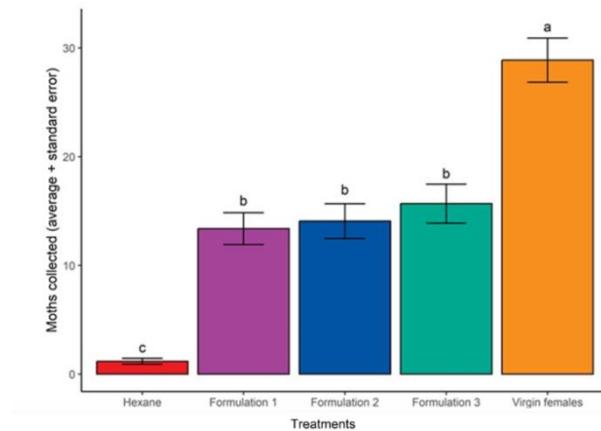


Figure 2. Number of *Spodoptera frugiperda* individuals collected in each of the treatments in the cotton crop in the 2020/2021 harvest in Mato Grosso.

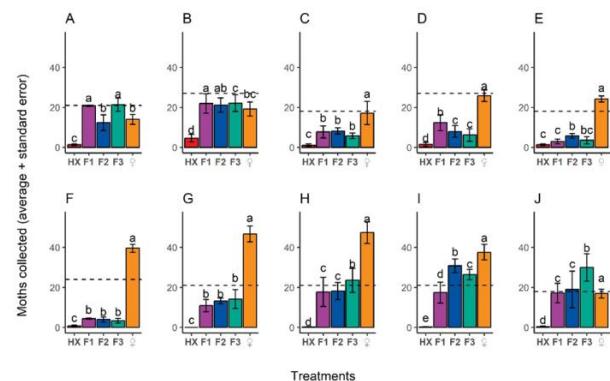


Figure 3. Number of *Spodoptera frugiperda* individuals collected, weekly, in each treatment in the cotton crop in the 2020/2021 harvest in Mato Grosso. HX: hexane (negative control); F1: formulation 1 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac); F2: formulation 2 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac); F3: formulation 3 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac + E2-6: Al); ♀: virgin females; * Different letters indicate significant difference ($p < 0.05$), in GLM and Tukey test; ** Dotted lines indicate the Level of Control adapted from Cruz et al. (2012).

From the third week of evaluation, a gradual loss of attractiveness of the formulations was observed, which continued until the sixth week (Figure 3C-E), when the septum was changed, therefore, in this period, only the virgin females reached the level of control. This period also coincided with the application of insecticides in the area, with emphasis on the evaluations of 05/18 (bifenthrin and

carbosuphane + phenylthiourea + strobilurin and triazole + etoxazole) and 06/03 (bifetin and carbosuphane + zeta-cypermethrin), which showed a reduction in the collection of moths in the formulations (Figure 4).

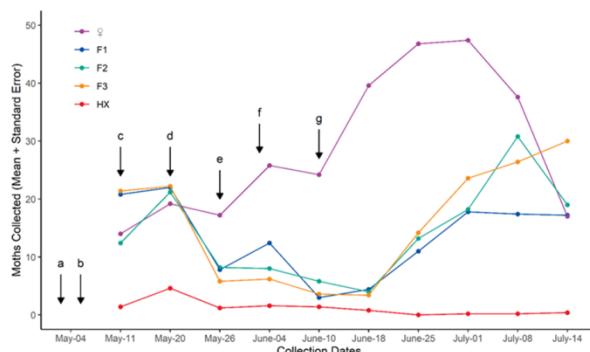


Figure 4. Population fluctuation of *Spodoptera frugiperda* collected in pheromone traps, contrasted with applications of phytosanitary products in cotton culture during the 2020/2021 season in Mato Grosso a: Malathion insecticide (organophosphate); b: Score fungicide (triazole) + Talisman insecticide (pyrethroid and carbamate); c: Polytrin insecticide-acaricide (organophosphate and pyrethroid) + Fungicide Score (triazole); d: Talisman insecticide-acaricide (pyrethroid and carbamate) + Polo insecticide-acaricide (phenylthiourea) + Priori top fungicide (strobilurin and triazole) + Smite insecticide (etoxazole); e: Fury insecticide (pyrethroid); f: Talisman insecticide-acaricide (pyrethroid and carbamate) + Fury insecticide (pyrethroid); g: Polytrin insecticide-acaricide (organophosphate and pyrethroid) + Polo insecticide-acaricide (phenylthiourea). HX: hexane (negative control); F1: formulation 1 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac); F2: formulation 2 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac); F3: formulation 3 (Z9-14: Ac + Z11-16: Ac + Z7-12: Ac + E2-6: Al); ♀: virgin females *Dotted line represents releaser replacement.

From the seventh week, the average capture grew progressively until the ninth evaluation, with oscillations in the capture of moths with the tested formulations (Figure 3G-I), and these reached the level of control in the eighth and ninth week, while the virgin females showed higher capture values in relation to the formulations during the seventh and ninth week. In the tenth week there was a reduction in the capture of moths between treatments, however, with virgin females the average number of moths captured was still close to the control level (Figure 3J).

Discussion

In the present study, the formulations containing Z9-14:Ac, Z11-16:Ac, and Z7-12:Ac proved effective in attracting *S. frugiperda* in cotton fields, confirming that these compounds function as key behavioural cues for this species under local conditions. The consistent male captures observed with these blends reinforce their biological relevance and also agree with previous reports that identified these substances as primary components of the pheromone system in North American populations (Unbehend et al., 2013; Lima & McNeil, 2009),

suggesting functional stability of attraction across geographically distinct regions.

Pheromone attractiveness can vary according to the amount incorporated into the septa, the proportion of components, and field conditions (Cruz-Esteban et al., 2020). However, in the present study, no significant differences in moth capture were detected among these three variables. Contrary to the assumption that higher dosages enhance attractiveness, the incorporation of 6 mg into the septa, with or without the addition of the foliar compound E2-6:Al, did not improve capture rates compared with 3 mg (Cruz-Esteban et al., 2020). This result is consistent with the findings of Cruz-Esteban et al. (2020), who reported the highest capture efficiency for *S. frugiperda* at a dose of 600 µg (0.6 mg).

The concentrations evaluated in this study are within the range previously reported as effective for *S. frugiperda* monitoring, supporting the suitability of the tested dosages. In particular, the proportion of Z7-12:Ac used is consistent with recommendations for improving capture efficiency and may have contributed to the stable attractiveness observed among the formulations. In addition, the attractive period of approximately three weeks recorded for the septa is similar to that reported for comparable pheromone blends under field conditions. The persistence of the odour is directly related to the longevity of attractiveness, which may contribute to lower trap maintenance costs and reduced frequency of lure replacement (Cruz-Esteban et al., 2018; Cruz-Esteban et al., 2020; Unbehend et al., 2014; Bratovich et al., 2019; Melo et al., 2011).

Despite agreement with previous studies, the lower efficiency of the synthetic pheromone formulations compared with virgin females remains unexplained. The most plausible explanation is geographic variation arising from reproductive isolation and/or the occurrence of distinct *S. frugiperda* strains in different regions (Cruz-Esteban et al., 2020; Muthukumar & Kennedy, 2021; Unbehend et al., 2013, 2014).

The synthetic compounds tested in this study, although selected based on data from the Pherobase, were less attractive than virgin females under field conditions. This reduced performance suggests that local populations may respond differently to specific pheromone blends, which is consistent with reports of geographic and strain-related variation in *S. frugiperda* populations (Unbehend et al., 2013, 2014). Such variation could explain the limited effectiveness observed in this study, even when using components considered standard for the species.

These findings underscore the necessity for more detailed studies involving strains from Mato Grosso to isolate and identify pheromonal compounds from local populations, thereby improving pest monitoring efficiency. This is crucial given the importance of the tool for management because, despite repeated applications of insecticides, the numbers of moths collected above the control limit of three moths per trap as proposed by Cruz et al. (2012).

Conclusions

This study demonstrated that the tested synthetic pheromone formulations can attract *Spodoptera frugiperda* in cotton fields, confirming their potential application as monitoring tools under the conditions of Mato Grosso. However, their performance remained inferior to that of virgin females, indicating that the current blends do not yet fully reproduce the attractiveness of the natural pheromone. This finding suggests the influence of geographic or population-specific variation and highlights the need for regional optimisation of pheromone composition. The persistence of attractiveness over several weeks supports the technical feasibility of their use in field monitoring programmes. Overall, the results contribute to the development of more efficient and locally adapted semiochemical-based strategies for the integrated management of *S. frugiperda* in cotton agroecosystems.

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Interests conflict

The authors declare that they have no conflict of interest. The research was conducted without financial support from external sources and in the absence of any commercial or financial relationships that could be considered potentially conflicting with the results presented.

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