STINK BUGS INCIDENCE ON BT COTTON IN BRAZIL

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Abstract

The economic impact evaluation of the boll-feeding stink bugs complex on Bollgard® cotton becomes necessary for the validation of the benefits from this technology in the cotton production systems in Brazil. This work studied the population of soybean migrant stink bugs species on cotton varieties (Bt and non-Bt) with the same parental, cultivated in an area bordered by a soybean crop, at the 22º 11' of latitude South and 54º 56' of longitude West, in Dourados, in the state of Mato Grosso do Sul, Brazil. The sources of variation variety (Bt and non-Bt), Nodes Above White Flower – NAWF (6-7, 5-6, 4-5, 3-4 e < 3) and interaction between variety and NAWF were tested to the numbers of nymphs, adults and total of specimens (adults + nymphs) of stink bugs species observed per shake sheet. At the end of the growing season, the cotton seed and lint yield were evaluated. Three stink bugs species occurred at the five cotton plants reproductive phases: *Euschistus heros* (Fabr., 1798), *Edessa meditabunda* (Fabr., 1794) and *Nezara viridula* (L., 1758). No differences to the stink bugs infestations for the sources of variation variety and interaction between variety and NAWF were detected. The most abundant species was *E. heros*, presenting mean number of adults per shake sheet higher since the 4-5 NAWF phase, coinciding with the full maturity phase of the soybean cultivated at the adjacencies of the experimental area. The colonization of the stink bugs species on the cotton varieties could be evidenced, due to the nymphs incidences throughout the evaluation period. Particularly, *E. meditabunda* seems to be the most adapted species to colonize the cotton. At the experimental conditions, the non-Bt variety presented means of seed and lint cotton yield significantly higher than the Bt variety, suggesting variation in the genetic composition of the varieties, although the Bt variety is considered essentially derived from the non-Bt.

Introduction

In Brazil, the Bollgard® cotton has been legally available since 2005 and it is until this moment the only technology of genetically modified insect resistant cotton regularized in this country. It is estimated that in the Brazilian growing season of 2007/2008, 6.7% of the cotton producer area was cultivated with Bt varieties (Monsanto, personal communication). It is possible that the area cultivated with Bollgard® increases, because the target-pests of this technology [*Heliothis virescens* (Fabr., 1781), *Pectinophora gossypiella* (Saund., 1844) and *Alabama argillacea* (Hueb., 1818)] are important pests of the cotton cultivated in the main cotton producer regions of Brazil.

At the Brazilian conditions, more than 20 species of insects can cause damages to the cotton, among which *H. virescens*, *Anthonomus grandis* Boh., 1843 and *Aphis gossypii* Glover, 1877 detach. Phytophagous bugs of the Pentatomidae family, insect-pests of occasional occurrence and secondary importance to the cotton in Brazil have occurred with high frequency and intensity on the Brazilian cotton fields at the latest growing seasons, mainly where long cycle varieties are cultivated and/or that receive less numbers of insecticide sprays (Papa, 2006).

This fact could be directly related to the migration of these species from the soybean at the end of its cycle to the cotton plants, since this leguminous occupies a huge extension of cultivated area in Brazil. Besides, stink bugs could be encountered in many host plants, cultivated or not, being able to migrate from one species to another looking for shelter and food (Panizzi, 1997).

In the Brazilian Center-West region where 60% of the cotton producer area is concentrated, the cotton crops divide space with soybean crops, representing only 6.8% of the total area cultivated with this two crops, being the stink bugs complex [*E. heros*, *Edessa meditabunda* (Fabr., 1794), *N. viridula*, *Piezodorus guildinii* (West., 1837), *Chinavia* spp. and *Thyanta perditor* (Fabr., 1794)] the main pest group of the soybean cultivated in this region. In these conditions, the stink bugs migration from soybean to cotton can occur once the soybean cycle is shorter than the cotton cycle and these two crops are cultivated at the same time and space. The end of the reproductive phase of the soybean crops still coincides with the full reproductive development (bolls formation, squares emission and bloom) of the cotton plants, period in which the plant is more susceptible to the attack of these insects (Greene et al., 2006).

Considering that the adoption of the Bt technology by Brazilian producers during the growing seasons increases, with the consequent reduction of the numbers of insecticide sprays on the target-pests of the technology, allied to the regional boll weevil suppression programs, reducing even more the numbers of insecticide sprays, and to the adoption of more selective and specific insecticides (or with reduced spectrum) to control the non-target pests; the stink bug complex could acquire economic importance to the cotton cultivated in Brazil, as in the Southeast and Mid-South of the USA since 1996, with the beginning of the Bt varieties cultivation and the boll weevil eradication (Haney et al. 1996; Roberts, 1999; Edge et al. 2001; Torres & Ruberson, 2005).

In this way, bioecologics and economic impact studies of the phytophagous stink bugs from the Pentatomidae family (migrants from soybean fields) on the cotton cultivated in Brazil become necessary for the establishment of integrated management strategies of these insects to the cotton production systems of Brazil.

This work aimed at evaluating the migrant stink bug complex from soybean at the end of its cycle on the Bt (Bollgard®) and non-Bt cotton, and the impact of these insects on the cotton yields from these cotton varieties in Brazilian conditions.

Materials and Methods

The trial was carried out during the growing season 2007/2008, in the experimental field of the Agricultural Science College, of the Federal University of Grande Dourados, in Dourados, in the state of Mato Grosso do Sul, Brazil, at 22° 11' of latitude South , 54° 56' of longitude West and 450 m of altitude.

The natural infestation of soybean migrant stink bugs was evaluated on two cotton varieties with the same parental, being one a Bollgard® variety (NuOpal®) and another a non-Bt variety (DeltaOpal®). Therefore, five evaluations of the stink bugs population, characterized by the Nodes Above the White Flower – (NAWF) (Oosterhuis et al. 1993) were made throughout the reproductive phase of the cotton plants. At the end of the cycle of the cotton plants the yield of each variety was determined.

The experimental area was installed in the center of an area of 60.000 m^2 which cultivate soybean without using insecticide sprays to control the stink bugs complex. A completely randomized experimental design was adopted with 12 replications, totalizing 24 experimental unities, in which each unity was compounded by 13 rows of the Bt or non-Bt variety, with 5 m in length, 0.9 m of row centers and presenting a density of 10 to 11 plants/m. The cotton varieties were seeding when the soybean cultivated around the experimental area was at the end of the vegetative development (38 days after the seedling emergence) (Fehr & Caviness, 1977).

The varieties were fertilized with 400 kg/ha of the NPK (08-30-20) fertilizer in the seeding line, and with 150 kg/ha of urea in two top dressing fertilizations, done on the 48th and 61st days after seedling emergence. The same production system was adopted for the two varieties and it follows the agronomics recommendations for the conventional cotton production used in Brazil.

To cancel the effect of other pests, systematic insecticide sprays were thrown every seven days in all experimental unities until fifteen days before the beginning of the stink bugs evaluations, to permit these insects to migrate from soybean to cotton, at the desired moment. After the evaluations, the sprays were thrown again and extended until the end of the cycle of the cotton plants.

Five evaluations using a white shake sheet with 1.0 x 0.9 m were made shaking vigorously two meters of parallels rows, in two random locals at the row centers of the 11 central rows of each plot. The stink bugs species observed were quantified and grouped in three categories: adults, nymphs and total (adults + nymphs). Each evaluation corresponded to one reproductive development stage of the cotton varieties and of the experimental area adjacent soybean, like the phenological scale proposal by Fehr & Caviness (1977) and Oosterhuis et al. (1993) to the soybean and to the cotton respectively. In this way, the 1st evaluation corresponding to the 6-7 NAWF stage to the cotton and to the R6-R7 stage to the soybean, the 2nd, to the 5-6 NAWF stage to the cotton and the R8 stage to the soybean, the 3rd, to the 4-5 NAWF stage to the cotton and R8 stage to the soybean and the 5th, to the < 3 NAWF to the cotton and R9 to the soybean (harvested soybean).

At the full maturity of the cotton plants, the seed cotton produced at the three central rows of each plot (13.5 m^2) was hand-picked and weighed in a precision balance to determine the seed cotton yield. One sample of 0.2 kg was removed from the total harvested in each plot, beneficiated, and the % of medium lint income of each variety was utilized for the lint yield estimation at the harvested area of each plot.

For the statistical analysis, the medium values of adults and nymphs of each stink bug species of each plot and evaluation were transformed in square root of (x + 0.5). The transformed data were analyzed based on the statistical methodology of Repeated Measures in Time, using the MIXED procedure of the statistical package SAS®, considering a fixed and a random model (Littel et al., 1998). The effects of varieties, phenological development (NAWF) and the interaction between varieties and phenological development (NAWF) were tested in the fixed model. Eight structures of covariance were tested for modeling the correlations between the repeated measures in the random model. The matrices tested were: Compound Symmetry (CS), Heterogeneous Compound Symmetry (CSH), First-Order Autoregressive [AR(1)], Heterogeneous AR(1) [ARH(1)], First-Order Autoregressive Moving-Average [ARMA(1,1)], Heterogeneous Toeplitz (TOEPH), Unstructured Correlations (UNR) and Variance Components (VC) (Malheiros, 2004).

Using the Corrected Akaike Information Criterion (AICC) the variance-covariance matrix was chosen to test the fixed effects (Burnham & Anderson, 2004). When the F test was significant ($P \leq 0.05$), the means were adjusted so as to be compared by the Tukey-Kramer test ($P \leq 0.05$), using the LS MEANS command from SAS®.

The data of seed and lint cotton yield of the varieties were submitted to the variance analysis, using the GLM procedure from SAS®. After evidencing the significance ($P \leq 0.05$) of the F test to the difference between varieties, the means of each variety were adjusted and compared by the Student's t-test ($P \leq 0.05$), using the LS MEANS and MEANS commands from SAS®.

Results and Discussion

The stink bug soybean migrant complex in the two cotton varieties throughout the evaluations was constituted by the species *E. heros*, *E. meditabunda* and *N. viridula*. Regardless of the sources of variation, the most abundant species throughout the evaluations period was *E. heros*, with 2168 specimens observed, followed by *E. meditabunda*, with 512 specimens, and *N. viridula*, with 115 specimens. None of the three species categories presented significance ($P \leq 0.05$) to the sources of variation variety and interaction between variety and NAWF. However, all species and categories were significant ($P \leq 0.01$) to the source of variation NAWF (Table 1).

In the 4-5 NAWF phase (moment when the soybean cultivated around the experimental area was at the R8 full maturity stage), *E. heros* adults, *E. meditabunda* nymphs and the total of specimens (adults + nymphs) of *E. heros* and *E. meditabunda* had presented means of individuals per shake sheet significantly higher than the previous evaluations, without significant reduction of these means in the posterior evaluations. The *N. viridula* had presented means of individuals per shake sheet significantly higher than the previous evaluations since the 3-4 NAWF phase and the adults of *E. meditabunda* and *N. viridula*, the *E. heros* nymphs and the total of specimens (adults + nymphs) of *N. viridula*, only in the last evaluated phase (< 3 NAWF) (Table 2 and Figure 1).

The results show evidences of the stink bugs migration phenomenon from soybean to cotton when this leguminous is closer to the full maturity stage, and suggest among the three observed species that *E. meditabunda* seems to be the most adapted species to colonize the cotton, considering that the significant growth of the mean number of nymphs per shake sheet for this species had occurred since the 4-5 NAWF phase and the mean number of *E. heros* nymphs species that had presented significantly increase since the 4-5 NAWF phase too was statically higher than the other means only at the last evaluation (< 3 NAWF). Another fact is that the means of *E. meditabunda* nymphs were numerically higher than the means of *E. heros* nymphs in the last three evaluations.

Although no significant differences had been detected to the sources of variation variety and interaction between variety and NAWF in none of the observed stink bugs species categories, the seed and lint cotton yield was statistically different between varieties (Figures 2 and 3).

Table 1. Significance levels of the tested sources of variation to the natural infestation of three migrant stink bugs species on cotton. Dourados, MS, Brazil. Growing season 2007/2008.

Source of Variation	Adult F	Adult *P*	Adult S.C.[1]	Nymph F	Nymph *P*	Nymph S.C.	Adult + Nymph F	Adult + Nymph *P*	Adult + Nymph S.C.
Euschistus heros									
Variety[2]	0.00	0.9624$^{NS(5)}$		0.79	0.3833^{NS}		0.37	0.5489^{NS}	
NAWF[3]	192.92	< 0.0001$^{**(6)}$	TOEPH	10.54	< 0.0001**	UNR	212.71	< 0.0001**	ARH(1)
Variety x NAWF	0.30	0.8804^{NS}		0.39	0.8166^{NS}		0.46	0.7639^{NS}	
C.V.[4] (%)		18.17			33.02			14.04	
Edessa meditabunda									
Variety	1.47	0.2385^{NS}		2.87	0.1045^{NS}		3.58	0.0716^{NS}	
NAWF	15.04	< 0.0001**	CSH	41.02	< 0.0001**	ARH(1)	56.70	< 0.0001**	ARH(1)
Variety x NAWF	0.55	0.6997^{NS}		1.64	0.1717^{NS}		1.81	0.1333^{NS}	
C.V. (%)		30.06			21.68			16.05	
Nezara viridula									
Variety	0.84	0.3702^{NS}		1.24	0.2776^{NS}		2.44	0.1322^{NS}	
NAWF	16.77	< 0.0001**	CS	9.10	< 0.0001**	CS	25.93	< 0.0001**	AR(1)
Variety x NAWF	0.27	0.8979^{NS}		1.19	0.3205^{NS}		1.34	0.2627^{NS}	
C.V. (%)		21.99			24.59			16.03	

(1)S.C. = Structure of Covariance, where TOEPH = Heterogeneous Toeplitz, UNR = Unstructured Correlations, ARH(1) = Heterogeneous AR(1), CSH = Heterogeneous Compound Symmetry e CS = Compound Symmetry. (2)Two levels: Bt (NuOpal®) and non-Bt (DeltaOpal®). (3)NAWF = Nodes Above White Flower; five levels: 6-7, 5-6, 4-5, 3-4 e < 3. (4)C.V. = Coefficient of Variation. (5)NS = Nonsignificant ($P \leq 0.05$). (6)** = Significant ($P \leq 0.01$).

Table 2. Mean number (±SE) per shake sheet of migrant stink bugs from soybean in five reproductive phases of the Bt and non-Bt cottons (n=24). Dourados, MS, Brazil. Growing season 2007/2008.

Stink bugs		NAWF[1] 6-7	5-6	4-5	3-4	< 3
Euschistus heros	A[2]	0.86[5] ± 0.04d[6]	1.50 ± 0.12c	3.46 ± 0.12ab	3.91 ± 0.11a	3.39 ± 0.12b
	N[3]	0.78 ± 0.04c	0.96 ± 0.09bc	1.15 ± 0.08ab	1.14 ± 0.08ab	1.42 ± 0.10a
	A + N[4]	1.64 ± 0.05c	2.46 ± 0.14b	4.61 ± 0.20a	5.05 ± 0.14a	4.81 ± 0.14a
Edessa meditabunda	A	0.80 ± 0.03c	1.05 ± 0.08bc	1.21 ± 0.08ab	1.09 ± 0.07b	1.54 ± 0.11a
	N	0.72 ± 0.02b	0.92 ± 0.07b	1.34 ± 0.08a	1.47 ± 0.09a	1.45 ± 0.07a
	A + N	1.52 ± 0.03b	1.97 ± 0.11b	2.55 ± 0.13a	2.56 ± 0.14a	2.99 ± 0.11a
Nezara viridula	A	0.71 ± 0.03c	0.71 ± 0.03c	0.81 ± 0.03bc	0.92 ± 0.03ab	1.06 ± 0.03a
	N	0.71 ± 0.04b	0.71 ± 0.04b	0.75 ± 0.04b	0.92 ± 0.04a	0.96 ± 0.04a
	A + N	1.42 ± 0.05c	1.42 ± 0.05c	1.56 ± 0.05c	1.84 ± 0.05b	2.02 ± 0.05a

(1)NAWF = Nodes Above White Flower. (2)A = Adult. (3)N = Nymph. (4)A+ N = Adult + Nymph. (5)Means obtained through the original data transformed in square root of (x + 0.5). (6)Means followed by the same letter in lines do not have differences between each other by the Tukey-Kramer test ($P \leq 0.05$).

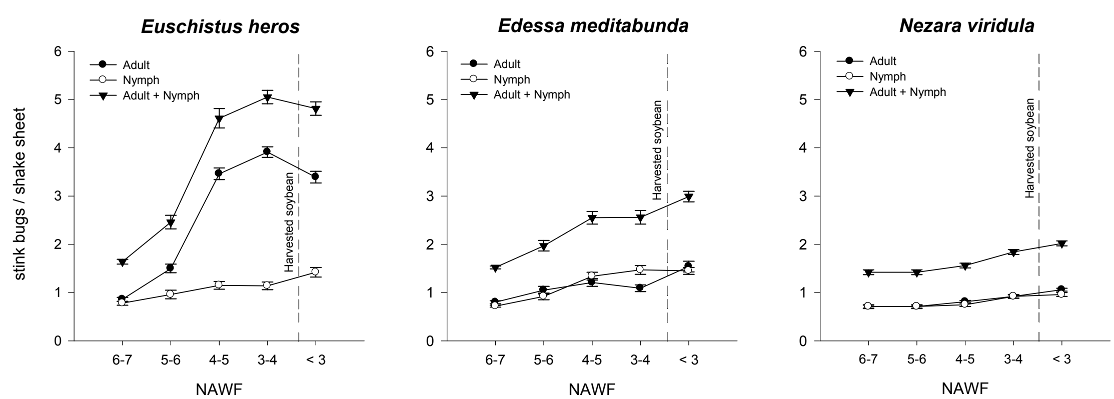

Figure 1. Population dynamic of three migrant stink bugs species throughout five phases of the reproductive development of the Bt and non-Bt cottons (n=24). Means (±SE) obtained through the original data transformed in square root of (x + 0.5). Dourados, MS, Brazil. Growing season 2007/2008.

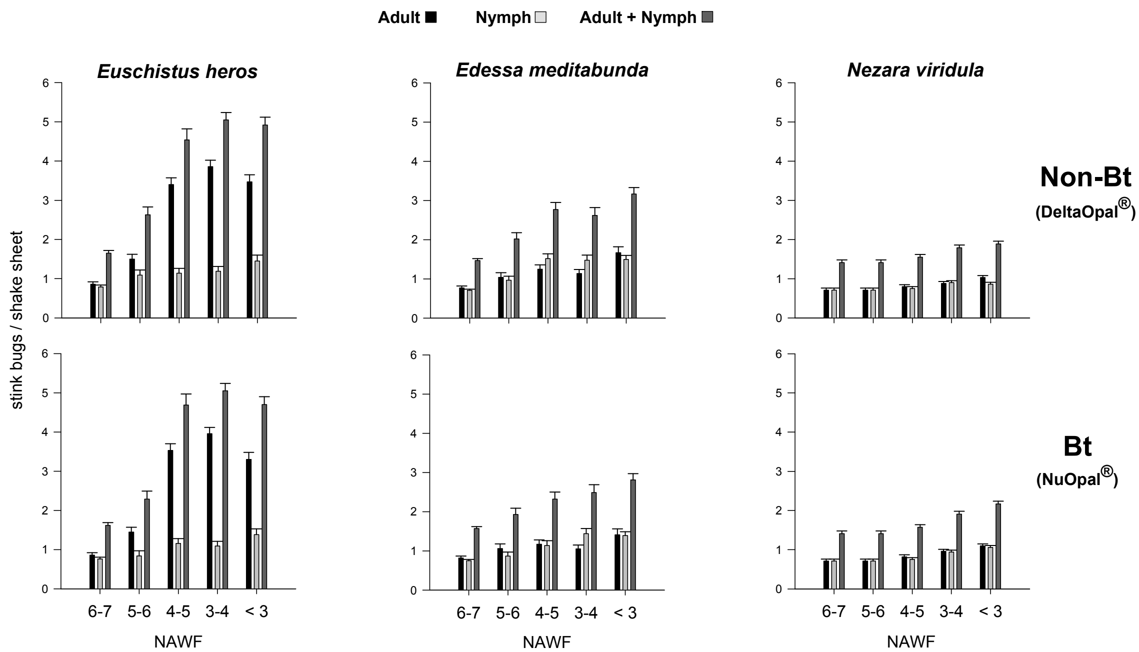

Figure 2. Population level of three migrants stink bugs species of soybean in five phases of the reproductive development of the Bt and non-Bt cotton (n=12). Means (±SE) obtained through the original data transformed in square root of (x + 0.5). Dourados, MS, Brazil. Growing season 2007/2008.

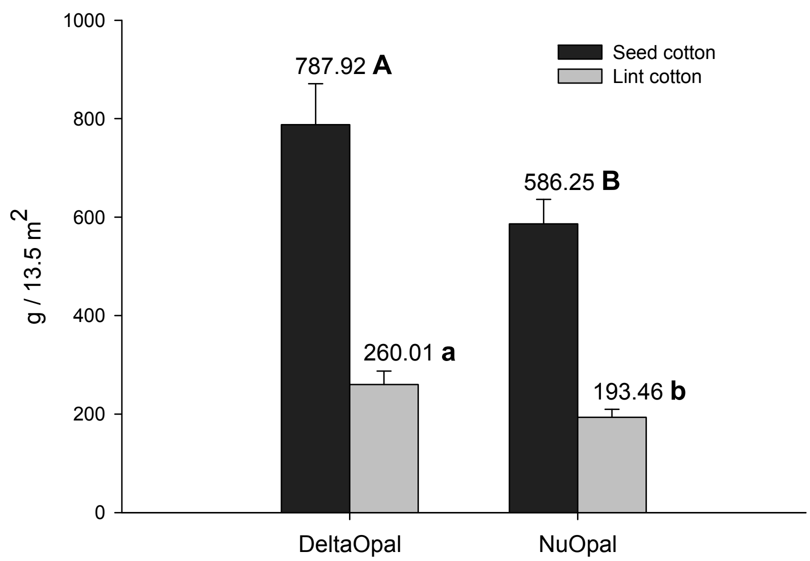

Figure 3. Mean yield of the non-Bt (DeltaOpal®) and Bt (NuOpal®) cotton varieties naturally infested by migrant stink bugs of soybean in five reproductive phases (n=12). Means (±SE) with the same letter within the same parameter do not have differences between each other by the Student's t-test ($P \leq 0.05$). Dourados, MS, Brazil. Growing season 2007/2008.

The tested varieties in this work have the same genetic composition (gene pool), with the yield and lint quality potential of the Bt variety being considered equal or superior to the non-Bt variety (Canci, 2007). However, what has been noticed in the same stink bugs infestation conditions is that the non-Bt variety showed to be more productive. This fact raises hypothesis about the real derivation existence of the non-Bt variety by the Bt variety or about the genetic comportment changes when the Bt variety is submitted to an adverse unexpected condition to the Brazilian reality, like a high density stink bugs infestation.

It is suggested that among other factors, the cultivated variety type could influence in the stink bugs damages and consequently in the fields yields (Roberts, 2006).

Through the nymphs incidences, the stink bugs colonization on the two types of cotton was demonstrated. The nymphs are not capable of migrating to cotton, but when the adults migration occurs and they oviposits, the nymphs could feed on the pinhead squares, favoring the abscission of these reproductive structures and interfering in the yield potential of the cotton plants (Willrich et al., 2004a).

The stink bugs migration from soybean varieties with different maturity cycles to the Bt and non-Bt cotton varieties NuCOTN 33b® and DPL 5415® was observed from the beginning of bloom to the beginning of bolls development, with stink bugs population peaks occurring throughout the period of bolls development, without significant yield differences between varieties (Bundy & McPherson, 2000). In the same way, on three Bt varieties, 4691B®, 4892BR® and BXN49B® was evidenced the same productive and quality yield performance when these varieties were submitted or not to stink bugs control (Bauer et al., 2006).

As the *E. heros* occurrence was the highest it is to expect that most of the boll damages has been caused by this species. In the case of *Euschistus servus* (Say, 1832), the seed cotton yield obtained from exposed bolls to an infestation with 30 adults/6.70 m^2 in the 6-9 and ≤ 3 NAWF phases was significantly lesser than the yield obtained from the non-infested bolls (Willrich et al., 2004b). The stink bugs scouting in order to define the most adequate moment for its control must be initiated by the bolls development phase (Willrich et al., 2004a).

This shows that the stink bugs scouting at the Brazilian cotton fields throughout the boll development period is essential, mainly when there is mature soybean closer to the cultivated area, as the stink bugs infestation is potential in the periods when the cotton plant is more susceptible to the stink bugs attacks.

Hence, more studies throughout the growing seasons that search the bioecologic comprehension of the stink bugs at the cultivated Bt and non-Bt cottons in Brazil are necessary to determine and validate the integrated management tactics of this pest complex at different brazilian cotton production systems, as soon as the infestation and damage potential of these insects at this experimental conditions were evidenced.

Conclusions

The stink bugs complex that was observed throughout the five phases of the reproductive stage of the Bt and non-Bt cotton plants was constituted by the species *E. heros*, *E. meditabunda* and *N. viridula*, being *E. heros* the most abundant species.

The maturation period of the soybean that is cultivated at the adjacencies of cotton fields can influence the migration of the stink bugs complex from soybean to cotton.

The infestation of the three incident stink bugs species did not present significant differences between varieties, despite the cotton plants colonization.

The non-Bt variety yield was significantly higher than the Bt variety at the same stink bug infestation conditions caused by the migration of the cultivated soybean at the adjacencies.

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