

BETWEEN-SEASON ATTRACTION OF COTTON BOLL WEEVIL, *Anthonomus grandis* Boh. (Coleoptera: Curculionidae) ADULTS BY ITS AGGREGATION PHEROMONE

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ABSTRACT: The present study was undertaken to investigate the attractiveness of boll weevil adults by its aggregation pheromone under winter field conditions. Two experimental fields were utilized at "Casa Branca", SP, Brazil. For each one, three areas were established near the refuge vegetation, separated 500 m from each other. Each area was divided in three sub-areas or blocks of 100 m² to receive pheromone applications (2.5 g per block). In addition to the pre-application counting, five additional evaluations were carried out after the pheromone application. Ten randomized sampling points per block were considered in each evaluation process. *A. grandis* adults responded immediately to the pheromone applications, and were captured for 14 days. The highest level of attractiveness was observed 24 hours after application. The application of the boll weevil aggregation pheromone during winter could increase the predation by natural enemies, due to the increase of prey availability. Chemical control can be recommended 24 hours after pheromone applications in small plots as a between-season strategy for the suppression of boll weevil adults.

Key words: *Anthonomus grandis*, Insecta, cotton, management

ATRAÇÃO DE ADULTOS DO BICUDO DO ALGODOEIRO, *Anthonomus Grandis* Boh. (Coleoptera: Curculionidae) POR SEU FEROMÔNIO DE AGREGAÇÃO NA ENTRESSAFRA

RESUMO: O presente estudo teve como objetivo investigar a atratividade do bicudo do algodoeiro ao seu feromônio de agregação em período de inverno. Foram utilizados dois campos experimentais no município de Casa Branca, SP. Em cada um destes, foram estabelecidas três áreas separadas de aproximadamente 500 metros, sempre próximas à vegetação de refúgio. Cada área foi dividida em três sub-áreas ou blocos de 100 m² para receber a aplicação do feromônio (2,5 g por bloco). Foi realizada uma contagem de bicudos adultos no solo antes, e mais cinco após a aplicação de feromônio. Em cada bloco, foram observadas dez parcelas aleatórias, para a avaliação dos bicudos. Os adultos de *A. grandis* foram atraídos imediatamente após a aplicação do feromônio, sendo capturados por mais de 14 dias após. O índice mais elevado de atração foi observado 24 horas após a aplicação. A utilização do feromônio de agregação durante o inverno, pode aumentar a ação reguladora exercida pelos inimigos naturais, devido ao aumento de disponibilidade de presas. Controle químico pode ser recomendado 24 horas após a aplicação de feromônio em pequenas parcelas, como uma estratégia para a supressão de adultos de *Anthonomus grandis* em período de entressafra.

Palavras-chave: *Anthonomus grandis* Insecta, algodão, manejo, entressafra

INTRODUCTION

The cotton boll weevil (BW), *Anthonomus grandis* Boheman, was detected in the São Paulo State, Brazil, for the first time in 1983 (Habib & Fernandes, 1983), and at the present time it is considered as one of the most important key-pests in Brazilian cotton fields. Many investigations were undertaken to study the population behavior, biological responses and other ecological aspects of this curculionid pest in this new habitat, to establish IPM strategies for its control (Fuchs & England, 1989; Gutierrez et al., 1991; Gabriel et al., 1992; Pierozzi Jr. & Habib, 1992a, 1992b, 1993a, 1993b; Ramalho & Silva, 1993; Fernandes et al., 1994; 1996; Araujo &

Azevedo, 1997). Many ecological and behavioral differences were observed among boll weevil populations in their new environment in comparison to other cotton agroecosystems, mainly of the U.S.A. (Pierozzi Jr., 1989).

Among several types of insect adaptations to environmental changes, some serve as protection against the seasonal climatic changes. Periodic migrations, diapause or dormancy are some of such adaptations. Regarding adults of the cotton boll weevil, since Brazzel Jr. & Newson (1959) described and characterized the diapause process, many other scientists have studied these adaptation mechanisms and their role in population dynamics. The spark of the diapause process seems to be associated to the limited availability of square buds

for the BW, low temperatures and plant exposition to short day length (Pierozzi Jr., 1989). The utilization of fruits (bolls) for larval boll weevil feeding is also considered as an important factor inducing diapause (Lloyd et al., 1967; Lloyd, 1986). Diapause, as well as dispersion, can be caused by some chemicals present in such fruits. According to Arellano (1995), the decrease in the amount of cotton squares and increase of boll availability can be considered as indicators of the winter period proximity and, consequently, the activation of the preparatory metabolism in the insect population. Increase of some metabolic rates, accumulation of fat tissues and drastic reduction of sexual and reproductive activities are some of the observed responses.

A facultative diapause occurs among adults of this curculionid pest. In the USA diapause is a survival mechanism of such a pest between the cotton seasons. This behavior occurs when the cotton season is completed, and especially when soil moisture decreases inducing a plant stress. This phenomenon is observed in Mexican strains as much as the US southwestern ones. In agreement with Pierozzi Jr. & Habib (1992a) we also believe that the boll weevils found in Brazil are of the same strain which occurs in Mexico and southwestern USA. According to Cross (1973), after the migration process, the diapausing weevils, and apparently some nondiapausing ones, dispersed into the forest litter for hibernation, and this type of dispersion occurs in regions with wet and cold between-season conditions. In other regions with dry and hot conditions during this period, dispersion would normally occur in order to localize an alternative host. Finally, some weevils would survive in old cotton stalks. Some authors consider the possibility that a part of *A. grandis* population does not accomplish the diapause process (Palmer & Cate 1992; Fernandes et al., 1994).

While BW growth and reproduction are not so common during overwinter seasons of temperate zones, and the diapause is vital for their populations; in the tropics the climatic conditions, including temperature and photoperiodism, are not sufficient to trigger this process. Moreover several host plants can stay available during the whole year offering pollens to feed adults (Tauber et al., 1986; Brown & Phillips Jr., 1989; Benedict et al., 1991; Chandler & Wright, 1991; Jones et al., 1992). Habib et al. (1984) captured great amounts of *A. grandis* adults in grandlure pheromone traps during winter.

In the Paraná State, southern Brazil, the presence of *A. grandis* as a key pest increased the cotton production costs during the last five years due to the number of insecticide applications which increased from an average of 5.4 to 10.1 applications per season (Morales et al., 1997). According to Mafra-Neto & Bakers (1996), two methods are used to prevent mating, using synthetic pheromones: "mating disruption" and "attract-and-kill". In the second method, pheromone

sources are mixed with small amounts of insecticides that poison the males and females attracted by the pheromone.

The present study was conducted to investigate the attractiveness of *A. grandis* adults to the aggregating pheromone (grandlure), under winter field conditions, in southwestern Brazil.

MATERIAL AND METHODS

Attractiveness bioassays were carried out during two consecutive between-season periods, at Casa Branca Municipality, SP, southeastern Brazil. For the first period, the study was undertaken in a cotton field (400 ha area, submitted to an IPM program), surrounded by soybean, corn and chrysanthemum fields in addition to a small secondary natural forest. For the second period, a 70 ha area also subjected to the same IPM program and surrounded by corn, sugar cane and orchard areas, was used. In both situations, the IPM program included the utilization of vegetable traps formed by cotton plants cultivated a month before on the boundary rows of the farm, which corresponded to about 5% of the total area. The cotton plant stalks were cut and eliminated as soon as the cotton was harvested, but after three months some sprouts could be observed in all area.

For each between-season period, three areas (A1, A2 and A3 with 500 m distance between each other) were established, near natural refuge vegetation or orchard areas, to study the pheromone attractiveness. Each area was divided in three separate blocks (100 m² each), 10 m apart from each other, and the first of them was 20 m away from the edge of the refuge area (Figure 1).

For each block, 10 randomized sampling points (1 m² each) were established for counting of the attracted boll weevil adults. One pre-application and five post-application counting (2 hours, 1, 7, 14 and 21 days) were carried out. In each block area, 2.5 g of grandlure pheromone (Blockaide™) were homogeneously distributed. Sites where BW adults were observed in each block (if flying, on the ground, on weeds or on cotton sprouts) were also observed. Statistical analysis was carried out with transformed data in $\sqrt{x+0.5}$, subjected to analysis of variance and Duncan's multiple range test.

RESULTS AND DISCUSSION

Before the "grandlure" application boll weevil adults were concentrated mainly in the refuge areas, in both periods of study. During the pre-application counting, no adults were found in the nine blocks of the first between-season period, while only two adults were encountered in the nine blocks of the second period. The little amount of sprouts, present in the bioassay area, were not enough to attract the over-wintering boll weevil

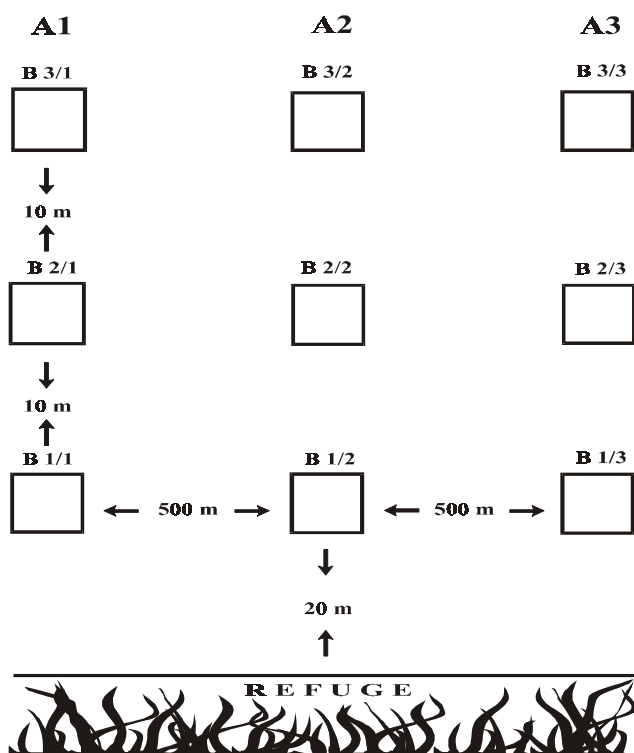


Figure 1 - Distribution scheme of areas and blocks in relation to the natural refuges.

adults. However, as shown in TABLE 1, they began to be attracted as soon as the pheromone was applied. The highest attraction was observed 24 hours after grandlure application in both between-season periods. Such attractiveness can be explained by the highest pheromone release and consequently the highest boll weevil response at the first days after release. Habib et al. (1984) studying the aggregating attractiveness of Blockaide™, detected a gradual attraction increase up to the sixth day.

The attracted weevils moved initially to sprouts and buds, and fed on them intensively. Authors observed up to nine adults in an only bud square; and in several occasions adults were observed in copulation.

A part of the boll weevil population was found to be feeding on weed pollen. However, the longer the time after pheromone application, the smaller the population on weeds or/and ground, thus concentrating on cotton sprouts and buds (TABLES 2 and 3).

Pheromone attractive quality and insect behavior were also studied among other coleopteran species in relation to their aggregation pheromones (Roelofs, 1981; Lloyd et al., 1981). According to Tumlinson et al. (1969) and White & Rummel (1978), the males are the first individuals to be attracted to the kairomone produced by the cotton plants. These attracted males start feeding and immediately produce and liberate their aggregation pheromone, attracting consequently the adult females. Such a behavior could be studied in a future investigation with *A. grandis* adults. Kairomone is a substance produced or acquired by an organism that, when it

TABLE 1 - Total number and mean (\pm SE) of cotton boll weevil adults per sampling point before and after pheromone applications.

Counting	1 st PERIOD		2 nd PERIOD			
	Number of adults		Number of adults			
	Total	Mean/sampling point	Total	Mean/sampling point		
Pre-application	00	00	a	02	0.02 \pm 0.38	A
2 hours after	19	0.20 \pm 0.78	ab	81	0.90 \pm 1.52	B
24 hours after	75	0.83 \pm 1.64	b	136	1.51 \pm 1.61	C
7 days after	11	0.12 \pm 0.77	ab	24	0.27 \pm 0.77	AB
14 days after	00	00	a	16	0.17 \pm 0.82	A
21 days after	00	00	a	09	0.10 \pm 0.74	A

TABLE 2 - Total and percentage of boll weevils found on sprouts, weeds and/or on ground before and after pheromone application, during the first period.

Phase	NUMBER OF ADULTS					
	On cotton sprouts		On weeds		On ground	
	Total	%	Total	%	Total	%
Pre-aplic.	0	0	0	0	0	0
2 h after	11	57.89	4	21.05	4	21.05
24 h after	54	72.00	7	9.33	14	18.67
7 d after	7	63.64	2	18.18	2	18.18
14 d after	0	0	0	0	0	0
21 d after	0	0	0	0	0	0

TABLE 3 - Total and percentage of boll weevils found on sprouts, weeds and/or on ground before and after pheromone application, during the second period.

Phase	NUMBER OF ADULTS					
	On cotton sprouts		On weeds		On ground	
	Total	%	Total	%	Total	%
Pre-aplic.	2	100	0	0	0	0
2 h after	43	53.09	36	44.44	2	2.47
24 h after	97	71.32	37	27.21	2	1.47
7 d after	3	12.5	1	4.17	20	83.33
14 d after	16	100	0	0	0	0
21 d after	9	100	0	0	0	0

reaches an individual of another species, evokes in the receiver a behavioral or a physiological reaction that is adaptively favorable to it, but not to the emitter (Nordlund, 1981).

Cotton kairomones, during the growing season, attract boll weevils; but small sprout rates during an overwinter period, are not sufficient to attract a significant number of *A. grandis* adults from surrounded areas, as shown in the present work. For this reason, it was very evident that the applied pheromone, in such a situation, was successfully able to attract a great number of active boll weevil adults from refuge areas within few hours after "grandlure" application.

The total absence of boll weevil adults, observed 14 and 21 days after pheromone application in the first period, showed the drastic effect of rain, reducing the pheromone attractiveness. On the other hand, during the second period, under no raining conditions, the high attractive capacity the extended up to 21 days.

Higher boll weevil presence in blocks nearest to the contiguous areas (block 1) was also observed. Those weevils were firstly attracted to blocks near to the refuge and then they pass to other blocks. However, no significant differences were observed between the number of adults found in those blocks during the two observation periods (TABLES 4 and 5). Moreover, it was observed that the amount of 2.5 g of grandlure / 100 m² showed to be economic and sufficient to result in a good attractiveness.

Summy et al. (1993) under USA cotton field conditions, observed *A. grandis* adults to be attracted to pheromone traps nearest to cotton plants residues during between-season periods. Gabriel et al. (1992) in Brazil, found a reduced number of active BW on soil, during winter in areas near cotton fields. Habib et al. (1984) and Fernandes (1995) utilizing grandlure pheromone traps captured great amounts of BW in winter season.

The present authors believe that applications of the boll weevil pheromone during winter, could increase the predatory action by natural enemies, due to the increase of prey availability. The performance of predator ants like *Solenopsis*, *Pheidole* and others, can promote a population decrease of boll weevil adults. Fernandes et al. (1994) observed that the potential benefit of ants, removing over-wintering *A. grandis* adults on the ground, during the inter-cropping season, is mainly that of reducing the risk of high level infestation during the subsequent cotton growing cycle.

In spite of the extensive bibliography regarding the diapause of BW in United States and Mexico (Brazzel & Newson, 1959; Mitchel et al., 1973) and few studies in Brazil (Campanhola et al., 1986, Campanhola & Martin, 1987) it seems clear to the authors that, a high proportion of BW adults remains active under field conditions of southeastern Brazil, if cotton buds or alternative hosts plants, in adjacent areas, were available. Even those individuals with apparently diapause indicators, as atrophied reproductive organs, fat accumulation and

TABLE 4 - Mean (\pm SE) of Boll Weevils found in three blocks at the first observation period before and after pheromone application.

Counting	Boll Weevils founded		
	Block 1	Block 2	Block 3
Pre-aplic.	0.00 \pm 0.00	0.00 \pm 0.00	0.00 \pm 0.00
2h after	1.33 \pm 0.76	3.33 \pm 1.44	1.67 \pm 0.76
24 h after	5.33 \pm 2.75	13.67 \pm 4.11	6.00 \pm 2.46
7 d after	0.33 \pm 0.76	1.00 \pm 1.00	2.33 \pm 1.51
14 d after	0.00 \pm 0.00	0.00 \pm 0.00	0.00 \pm 0.00
21 d after	0.00 \pm 0.00	0.00 \pm 0.00	0.00 \pm 0.00

Difference between three blocks were not significant. F= 0,260 P>0,01, ANOVA test). Means transformed by $\sqrt{x+0.5}$.

TABLE 5 - Mean (\pm SE) of Boll Weevils found in three blocks at the second observation period before and after pheromone application.

Counting	Boll Weevil founded		
	Block 1	Block 2	Block 3
Pre-aplic.	0.67 \pm 0.76	0.00 \pm 0.00	0.00 \pm 0.00
2h after	13.67 \pm 2.65	5.33 \pm 1.79	8.00 \pm 3.08
24 h after	21.67 \pm 3.55	17.00 \pm 4.36	6.67 \pm 1.23
7 d after	4.67 \pm 1.07	2.33 \pm 0.76	1.00 \pm 0.00
14 d after	1.33 \pm 1.51	2.67 \pm 1.51	1.33 \pm 1.23
21 d after	1.67 \pm 1.70	0.33 \pm 0.76	1.00 \pm 1.00

Difference between three blocks were not significant. F= 0,585 P>0,01, ANOVA test). Means transformed by $\sqrt{x+0.5}$.

decrease of some metabolic activities, were observed to be only in a quiescent phase, but not in diapause, in agreement with Pierozzi Jr. & Habib (1993b). According to Tauber et al. (1986) quiescence is a reversible state of suppressed metabolism imposed by environmental conditions such as temperature, humidity and unfavorable nutrition. Guerra et al. (1984), studying boll weevils in tropical areas in south of Mexico, observed quiescent forms and not a real diapause. Similar observations were made by Brazzel & Newson (1959), who detected reproductive activities among the BW adults, during inter-cropping season.

Finally, it could be said that, at the end of the cotton season, the boll weevil adults disperse looking for alternative host plants, reproductive resources and refuge vegetations to spend the between-season period. Studies related with the permanence of boll weevils in refuge

areas on alternative host plants were carried out by Campanhola et al. (1986), Lukefahr et al. (1986), Brown & Phillips Jr. (1989) and Pierozzi Jr. & Habib (1993b).

The attraction of adults of *A. grandis* to the aggregation pheromone Grandlureä, in small areas, according to the results of the present research, could be considered as a benefit IPM strategy during winter. Chemical insecticide treatments in these trap blocks (attract and kill), could be recommended 24 hours after pheromone application, as a between-season control method to suppress *A. grandis* adults. This practice, certainly, would reduce the number of chemical insecticide applications and, consequently, the cost of the control of such a pest, during the growing season.

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