STIMULI CAPABLE OF ATTRACTING *Triatoma infestans* (KLUG, 1834) (HEMIPTERA: REDUVIIDAE) UNDER LABORATORY CONDITIONS

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RESUMO

Estímulos capazes de atrair *Triatoma infestans* (Klug, 1834) (Hemiptera: Reduviidae) sob condições de laboratório

Observou-se o comportamento de *Triatoma infestans* (Klug, 1834) quando submetidos a diferentes períodos de privação alimentar e colocados a diferentes distâncias do estímulo. Investigou-se o potencial do pombo e do camundongo como fontes de estimulo sobre os referidos insetos.

Os experimentos foram realizados em laboratório e os equipamentos utilizados constavam de caixas de escolha. Concluiu-se que: a) o tempo de privação alimentar está diretamente relacionado com a intensidade de deslocamentos ambulatórios, independentemente da presença de fontes de estímulo; b) as fêmeas têm mais resistência à privação alimentar que os machos; c) o pombo representa uma fonte de estímulo mais atuante que o camundongo; d) o camundongo atraiu os insetos somente quando colocado muito próximo deles.

INTRODUCTION

The studies of stimuli which attract triatomines normally report the existence of pheromones, substances that act at the intraspecific level and that are also important at the social

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organization level (ANTICH, 1965, 1968; BALDWIN *et al.*, 1971). However, the possibility that the food source itself may act as a source of stimulation for these insects has not been taken into account in studies on food habits, since identification is usually made on the basis of the contents of the stomach of triatomines (ROCHA E SILVA *et al.*, 1977), or by isoenzyme analysis (ALMEIDA, 1982a, b).

In general, among the factors that may affect the attraction of triatomines, it could be pointed out the distance from the source of stimulation (MINTER *et al.*, 1973) and the time of food deprivation of these insects (PERLOWAGORA-SZUMLEWICZ, 1952; RYCKMAN, 1952).

Even though BARRETTO (1958) has shown that the triatomine species found in human dwellings have eclectic food habits, MINTER (1976) has stated that *Triatoma infestans* (Klug, 1834) prefers feeding on man, and on birds and rodents after him. For this reason it was decided to use a pigeon and a mouse as sources of stimulation as a model in the present study.

The objective of the present study was to investigate the behavior of *T. infestans* with respect to sources of stimulation presented.

Using equipment of selection boxes the experiment was set out to determine whether: a) *T. infestans* is attracted by pigeons and mice; b) time of food deprivation affects attraction to pigeons and mice; c) distance from the source of stimulation affects attraction.

MATERIAL AND METHODS

The experiments were carried out using a set of selection boxes (Fig. 1) consisting of five boxes made of hard, smooth and transparent plastic and measuring 11 x 11 x 15 cm each. To decrease the amount of light penetrating into the boxes, they were painted with dull black paint on the outside. Lids were fitted under pressure to the top of each box, through which the animals used in the experiments were placed and removed. The internal, unpainted walls were smooth in order to prevent the insects from climbing them. The boxes were connected with rigid and opaque PVC tubing measuring 45 x 4 cm or 15 x 4 cm, depending on the distance of the source of stimulation. The ORIGIN box (a), in which the experimental animals were placed and located on a higher plane than the rest of the set, contained two side entrances closed by two small plastic windows (j) sliding on small tracks and which could

be opened vertically by pulling a wire (fa) connected to them and extending beyond the top of the box. The tubes were connected to these two entrances on one end and to two boxes, TRAP-1 (b) and TRAP-2 (c), on the other, in which the experimental animals could also be placed. These two boxes in turn were connected with two other ones (d-e) through two smaller pieces of tubing (6 x 4 cm). These were the STIMULUS boxes which were used for placement of the animals to be used as sources of stimulation. Seven independent sets of boxes were prepared and mounted on a 75-cm high table for easier handling. Lighting was provided naturally through the laboratory window, with no direct sunlight on the equipment. The temperature was maintained between 26 and 29°C with an air conditioner attached to a timer set at predetermined times, and relative air humidity was maintained at 85± 3%.

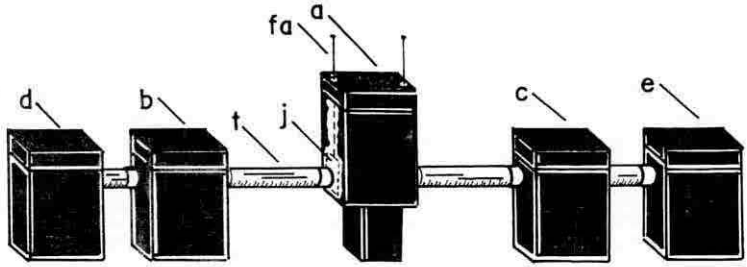

FIGURE 1 - Set of selection boxes. a: ORIGIN box; fa: wire; j: window; t: connecting tubes; b: TRAP-1 box; c: TRAP-2 box; d: STIMULUS-1 box; e: STIMULUS-2 box.

The selection boxes were used in the following experimental procedure:

1 - BASELINE

The experimental animals consisiting of triatomine pairs (10 males and 10 females), were placed in the ORIGIN box in the absence of any source of stimulation in the STIMULUS boxes.

2) NATURE OF THE SOURCE STIMULATION

The experiments between the experimental insects and the sources of stimulations were carried out in the selection boxes. The sources were a pigeon and a mouse.

3) DISTANCES FROM THE SOURCES OF STIMULATION

The sources of stimulation were tested at 75 and 45 cm from the experimental insects.

4) TIME OF FOOD DEPRIVATION

All triatomines were allowed to feed on pigeon blood until gorged and the experimental insects were then submitted to food deprivation at a sequence varying from 0 to 10 and 20 days.

5) CONTROL OF VARIABLES IN THE EXPERIMENTAL INSECTS

Control was done by the independent group plan (10 male and 10 female triatomines selected at random) with three replications for each assay, with a total of 30 couples for the baseline and 30 couples for each source of stimulation.

The experiments were always started at 8:00 a.m., when the experimental insects were placed in the ORIGIN box. This box was kept with the windows closed until 6:00 p.m., which represents the scotophase. At this time, the windows were opened, thus permitting the triatomines to move to the TRAP-1 or TRAP-2 box. On the following day, the windows were again closed at 8:00 a.m., the lids of the three boxes were removed and the insects were collected and counted.

The sources of stimulation were placed in the STIMULUS box according to the following criteria: the pigeon was immobilized inside the box by fixing the feet and wings close to the body with adhesive tape. The mouse was left unrestrained inside the box, but the orifice connecting with the TRAP box was obstructed with fine wire mesh to prevent its escape.

At the end of each test, the equipment was taken apart and washed with running water, liquid neutral detergent, 96 % alcohol and again with running water in order to eliminate any residues left by the animals and leave the boxes ready for use again.

FURTHER OBSERVATIONS

1) Pigeon and mouse stools were also used as stimulating sources. Since the results were negative (the experimental insects were not attracted), it was decided not to include them in the result tables.

2) The experimental insects, on the baseline, spread evenly through the TRAP-1 box, TRAP-2 box and ORIGIN box. Due to this fact, in the experiments between the experimental insects and the respective sources of stimulation, it was decided to place the pigeon and the mouse only in one side of the boxes, the STIMULUS-1 box, chosen at random. Based on the number of experimental insects detected in the boxes, statistical trials were applied (chi-square test), considering the subjects attracted or not by the stimulating sources.

3) In our pre-experiments, when aeration was applied inside the STIMULUS-1 box, the air pump caused a slight trembling, which hindered the movement of the experimental insects to that side. Due to this fact, it was decided not to use this device in the true experiments.

RESULTS AND DISCUSSION

1 - Baseline (Table 1)

The experimental insects submitted to food deprivation (FD) for 10 and 20 days, both males and females, were homogeneously distributed throughout ORIGIN, TRAP-1 and TRAP-2 boxes. When recently fed, however, most experimental insects remained in the ORIGIN box.

WIESINGER (1956), in a study of the ambulatory activity of *T. infestans*, concluded that the feeding status of the insect was one of the factors that induced this activity. BRADY (1975) reached a similar conclusion with fly *Glossina morsitans* West, showing a direct relationship between food deprivation and increased activity by the insect. In the present study, this fact supports the statement by SCHOFIELD & PATTERSON (1977) that triatomines tend to "rest" after feeding. Thus, this "rest" may represent an adaptative advantage for these insects, since the possibility of being stalked by predators during this period is lower.

TABLE 1 - Baseline - Experimental insects in the absence of a source of stimulation.

DISTANCE (cm)	DEPRIVATION (days)	MALES			FEMALES		
		TRAP 1	ORIGIN	TRAP 2	TRAP 1	ORIGIN	TRAP 2
75	0	4	21	5	5	20	5
	10	10	10	10	10	11	9
	20	11	09	10	11	10	9

2 - Tests involving the experimental insects and the sources of stimulation. (Table 2)

2.1 - Sources of stimulation: a PIGEON and a MOUSE respectively.

a) Experimental insects recently-fed: both male and female also showed no dislocation activity, with most of them indifferent to these sources of stimulation.

PERLOWAGORA-SZUMLEWICZ (1952), when observing the life cycle of *T. infestans* in the laboratory, adopted a meal schedule at five-day intervals, a time considered "sufficient for the insect to feel ready to bite again". RYCKMAN (1952), also studing *T. infestans* in the laboratory, recommended a feeding interval of 10 days. In the present study, it was decided to use a food deprivation schedule variyng from zero to 10 and 20 days, with the attracting sources (pigeon and mouse) at first place at a distance of 75 cm. The results show that the recently-fed triatomines were not attracted. Thus, when the distance was shortened to 45 cm, the use of recently-fed insects would not be necessary, just those with 10 and 20 days of starvation.

b) Experimental insects under food deprivation, in presence of the pigeon: males were attracted under both conditions of FD (10 and 20 days) and at both distances from the pigeon (75 and 45 cm). Females were attracted only after 20 days of FD and at both distances.

TABLE 2 - Origin of the source of stimulation. The values within parentheses are those expected according to the null hypothesis, and the X^2 values marked with a * indicate a significant difference at the 5% level.

			MALES				FEMALES			
SOURCE OF STIMULATION	DISTANCE (cm)	DEPRIVATION (days)	TRAP 1	ORIGIN	TRAP 2	X^2	TRAP 1	ORIGIN	TRAP 2	X^2
PIGEON	75	0	4 (4)	21 (21)	5 (5)	0	6 (5.5)	19 (19.5)	5 (5)	0.1
		10	17 (13.5)	11 (10.5)	2 (6)	7.19*	12 (11)	11 (11)	7 (8)	0.43
		20	19 (15)	7 (8)	4 (7)	5*	19 (15)	7 (8.5)	4 (6.5)	5*
	45	10	22 (11)	5 (7.5)	3 (6.5)	9.9*	8 (9)	13 (12)	9 (9)	0.37
		20	18 (14)	9 (9.5)	3 (6.5)	5.6*	20 (15.5)	6 (8)	4 (6.5)	5.02*
MOUSE	75	0	6 (5)	20 (20.5)	4 (4.5)	0.7	5 (5)	20 (20)	5 (5)	0.15
		10	12 (11)	7 (8.5)	11 (10.5)	0.71	10 (10)	9 (10)	11 (10)	0.2
		20	13 (12)	4 (6.5)	13 (11.5)	2.98	11 (11)	5 (7.5)	14 (11.5)	2.74
	45	10	17 (13.5)	10 (10)	3 (6.5)	5.8*	10 (10)	13 (12)	7 (8)	0.42
		20	9 (15)	8 (8.5)	3 (6.5)	6.1*	19 (15)	8 (9)	3 (6)	6.1*

SJOGREN & RYCKMAN (1966), in a field study on *T. protracta* (Uhler, 1894) observed that the males were more active than the females, and ESPINOLA (1973), in a laboratory study on *T. infestans* also observed the same phenomenon, as well as the fact that the activity of both males and females started during the scotophase period, between 6:00 and 7:00 p.m. MAC CORD *et al.* (1983) observed that the same insects, in the absence of an attracting stimulus (pigeon), showed very restricted activity starting at 9:00 p.m. on the first day and 7:30 p.m. on the second day. In presence of the attracting stimulus, the activity increased considerably starting at 1:00 p.m. on the first day and at 3:00 p.m. on the second day. Therefore, in the present study, even though the experimental insects had been placed in the ORIGIN box at 8:00 a.m., the windows of the box were opened only at 6:00 p.m., a time considered to be ideal. Thus, the insects were released into the adjoining boxes at the appropriate time and the time during which they were kept closed inside the ORIGIN box allowed them to acclimate to the environment.

The fact that males are more active than females may justify the results presented in Table 2, when insects submitted to 10 and 20 days of food deprivation moved preferentially towards the pigeon as a source of stimulation. The females, however, were attracted to the pigeon only after 20 days of food deprivation, regardless of the distance. This fact indicates that females are more resistant to food deprivation than males, suggesting that because of their reduced activity, their feeding physiology permitted a longer period of time to elapse until the next meal than in the males. This may represent an adaptative advantage for females.

c) Experimental insects under food deprivation, in presence of the mouse: experimental males were attracted under both conditions of FD (10 and 20 days) but only at 45 cm of distance from the mouse. Females were attracted only after 20 days of FD and only at 45 cm of distance from the mouse.

These results support the idea that females are more resistant to food deprivation than males.

According to WIESINGER (1956), temperature and carbon dioxide represent good attractents for *T. infestans*. During these experiments it was observed that when the pigeon and the mouse were removed from the respective boxes, these, as well as the animal's body, were covered with droplets of water. Since there was no forced aeration inside the boxes, the attraction phenomenon may have been due to the respiration and sweating of the stimulating sources, which, under the experimental conditions used, were quite intense.

The fact that attraction to the mouse occurred only when the experimental insects were at a distance of 45 cm can be

explained by taking into account the differences in proportionality between the body volume of the pigeon and the mouse, which may involve greater or smaller production of heat and carbon dioxide.

The fact that the pigeon acted as a better source of stimulation may be justified by the same considerations. However, MINTER *et al.* (1973), in a study involving the capture of *Panstrongylus megistus* (Burm. 1835) in human dwellings, suggested that these insects selected their hosts according to the distance.

In addition to the attractive caused by these sources of stimulation, triatomines themselves appear to exert this function intraspecifically by elaborating pheromones (BALDWIN *et al.*, 1971; NEVES, 1979) which shall be studied on furter experiments.

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ABSTRACT

The potencial of pigeons and mice as source of stimulation of *Triatoma infestans* was investigated by observing the behavior of these insects when submitted to different periods of food deprivation and placed at different distances from the stimulus.

The experiments were carried out in the laboratory using selection boxes. On the basis of the results, it was concluded that: a) the time of food deprivation is directly related to the intensity of ambulatory activity, regardless of the presence of stimulating sources; b) females are more resistant to food deprivation than males; c) pigeons are more effective source of stimulation than mice; d) The mouse attracted the experimental insects only when placed close to them.