

ON THE CAPTURE OF *TRIBOLIUM CASTANEUM* (HERBST): EFFECTS OF ALIVE AND DEAD INSECTS IN A TRAP

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ABSTRACT

The effectiveness of traps, whether baited or not baited with 4,8-dimethyldecanal (the pheromone of the red flour beetle *Tribolium castaneum* (Herbst)), is influenced by the presence of accumulated trapped specimens, living or dead.

The results of this study suggest that the presence of live insects in traps attracts other free specimens; however, under conditions of overcrowding quinone secreted by the insects regulates population density by counteracting the effect of the aggregation pheromone (4,8-dimethyldecanal). The quinone causes the insects to disperse or suppresses aggregation behaviour. The presence of dead insects in the traps, whether or not baited with 4,8-dimethyldecanal, can repel other free specimens of *T. castaneum*. This is probably due to the presence of a residual alarm pheromone produced by the insects before death.

INTRODUCTION

The aggregation pheromone of *Tribolium castaneum* (Herbst) is produced by the males at a continuous rate under all conditions. Quinones are secreted by both sexes but only under certain conditions, such as crowding, excitement, agitation and partial narcosis (Engelhardt *et al.*, 1965; Ogden, 1969; Irwin *et al.*, 1972). The aggregation pheromone and the quinones interact; the effects of this interaction depend on the number and sex of the beetles present in the infested commodity and on the duration of the infestation (Mondal, 1983, 1985, 1993; Mondal and Port, 1994). The quinones regulate the population density of *Tribolium* by counteracting the effect of the aggregation pheromone. Under conditions of overcrowding and lack of food, the secretion of aggregation pheromone on the male femora is inhibited, causing the insects to disperse or suppressing the aggregation behaviour (Faustini and Burkholder, 1987).

Use of the aggregation pheromone has been proposed in the monitoring and control of *Tribolium* spp. Where traps of various designs are used for monitoring this pest, pheromone traps appear to be more effective in the detection of low infestations (Burkholder and Ma, 1985; Mullen, 1992). The effectiveness of *Tribolium* pheromone-baited traps depends on many factors, including trap location, trapping duration, grain type and

condition, grain temperature, movement and dispersion of the insects and pheromone release (Fargo *et al.*, 1989; Barak *et al.*, 1990; Obeng-Ofori and Coaker, 1990a, b; Pinniger, 1990; Trematerra and Daolio, 1990; Trematerra, 1992; Trematerra *et al.*, 1996). An understanding of the environmental and physiological factors affecting insect response to pheromones is required for their successful employment in influencing insect behaviour. For this purpose a series of experiments was designed to measure the response of *T. castaneum* under different conditions. The main aim of the experiments was to determine if trap catches, whether or not the traps were baited with synthetic pheromones, were affected by the presence of accumulated living and dead insects.

MATERIALS AND METHODS

T. castaneum were cultured in a constant-environment room at $25 \pm 2^\circ\text{C}$ and 70% r.h. using wheat flour as the food medium.

The beetles were released and recaptured in two "Flit-Trak M" traps in a covered plastic cage ($43 \times 21 \times 10$ cm). One trap (A) was baited with cracked maize and *T. castaneum* adults at densities of 2, 5, 10, 20 and 40 live adults/trap, and the other trap (D) was baited with cracked maize and 10 dead insects. Fifty adult beetles of mixed sex and age were released into the cage. The experiment was carried out at $24 \pm 2^\circ\text{C}$.

The numbers of trapped *T. castaneum* were counted after 0.5, 1, 2 and 3 h, and again after 19 h, and the percentage frequency of detection was calculated. A set of all the counts under each of the experimental conditions was termed a 'watching run', and each 'watching run' was replicated four times.

A similar experiment using the same densities of *Tribolium* was carried out to determine the effect of the pheromone, 4,8-dimethyldecanal, on beetle-trap counts.

Insects found in traps baited with live insects were designated as "first choice," while insects found in traps baited with dead insects were termed "second choice." There was no guarantee that all the specimens attracted were trapped. Therefore, the number of specimens in the trap did not necessarily correspond to the number of specimens actually attracted during the watching time. Nevertheless, the specimens in each trap must be assumed to have chosen between the two available possibilities.

The presence of specimens under and over the traps can be attributed to partial failures in trap performance. Therefore the larger number of specimens in a given trap, reflecting the quantity of specimens which chose that trap, must indicate that the related data are reliable. According to such an assumption, the level of trap performance must be considered as a statistical weight.

On this basis, the efficiency or performance level of the first-choice trap is given by the ratio between the number of specimens actually captured in the trap and the number of specimens which chose it, i.e. specimens in, under and over the trap.

We assumed that the performance levels of the two traps were uncorrelated. We also assumed that the number of specimens was large enough so their independent response to the trap was a reliable indicator. On these bases, we define a mean efficiency of the

overall capturing performance as the arithmetical mean of the two levels of performance. We define this as overall capturing performance.

Three main possibilities (first choice, second choice or no choice) have to be accounted for for each given time interval and each experimental run. Of course the time required for the number of specimens to choose (or not to choose) a trap must also be analyzed.

As the experimental conditions change, the response of the group of specimens under examination also changes.

In principle, one could use the mean of these data. Unfortunately, this would not take into account the peculiarities, such as the differing reliability of the traps during the different runs, characteristic of each run.

Another point to take into account is the number of specimens which choose a trap in comparison with the overall number of specimens. Actually these two quantities are different and, in principle, the closer their ratio is to unity, the more reliable the trial is. Thus, the ratio between these two quantities can indicate the overall run reliability.

An interesting point is the time during which the specimens choose a trap (first or second choice) or do not choose. The statistical question is what percentage of specimens makes a choice during the time interval of the experimental run.

Thus the data in which one is interested emerge from the means of the rough experimental data weighted by overall capturing performance and overall run reliability. This is indicated by e_i (where i indicates time). It is of course also possible to do the same for non-choice, where the final data is indicated by e_i^* .

In principle one expects that $e_i + e_i^* = 1$, which is not the case since the data have been transformed. Therefore, the final data must be normalised in order to obtain the unity from the sum of these two quantities. Therefore the data we are looking for are:

$$e_i^{\text{norm}} = \frac{e_i}{e_i + e_i^*}; \quad e_i^{*\text{norm}} = \frac{e_i^*}{e_i + e_i^*}$$

Figures 1 and 2 show the issues for the variables $e_i^{*\text{norm}}$ and e_i^{norm} for the case with and without the pheromone for the two different experimental conditions, without pheromone in Fig. 1 and with pheromone in Fig. 2.

Another point to discuss is the distribution of live specimens between the first choice and the second choice. The rough data for analysis must be weighted by both the overall run reliability and the trap reliability. In this way, one obtains the quantities q^{first} and q^{second} . Once again, the data must be normalised to set the sum of the probabilities to unity. Therefore we get:

$$q^{\text{first (norm)}} = \frac{q^{\text{first}}}{q^{\text{first}} + q^{\text{first}*}} \quad \text{and} \quad q^{\text{second (norm)}} = \frac{q^{\text{second}}}{q^{\text{second}} + q^{\text{second}*}}$$

In Figs. 3 and 4 we report the behaviour of the quantities $q_i^{\text{A norm}}$ and $q_i^{\text{B norm}}$ as a function of i (time) under the two experimental conditions, with pheromone in Fig. 4 and without pheromone in Fig. 3.

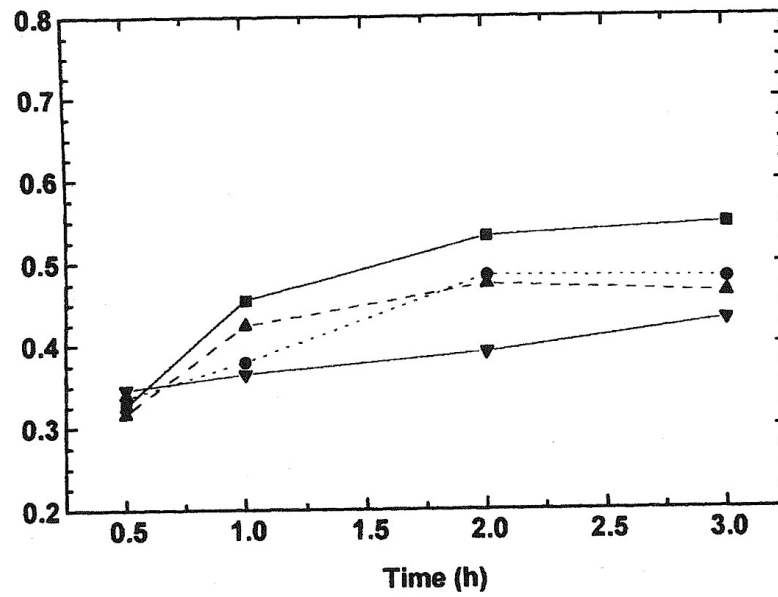


Fig. 1. Evolution in time of the variable e_i^{norm} (on the ordinate) when no pheromone is added. Squares: $A = 0$, $D = 10$; circles (dotted line): $A = 5$, $D = 10$; up triangles (dashed line): $A = 10$, $D = 10$; down triangles: $A = 40$, $D = 10$.

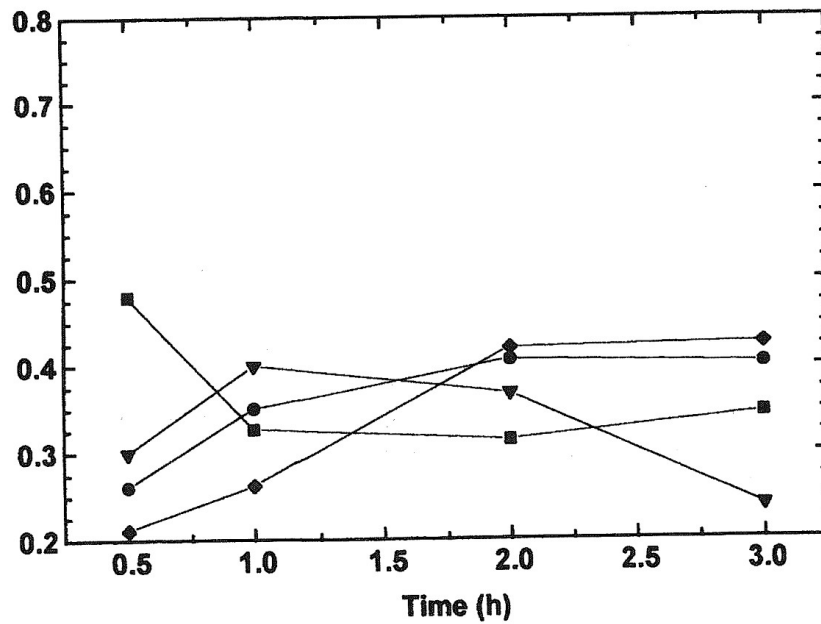


Fig. 2. Evolution in time of the variable e_i^{norm} (on the ordinate) when pheromone is added. Squares: $A = 0$, $D = 10$; circles: $A = 5$, $D = 10$; diamonds: $A = 20$, $D = 10$; down triangles: $A = 40$, $D = 10$.

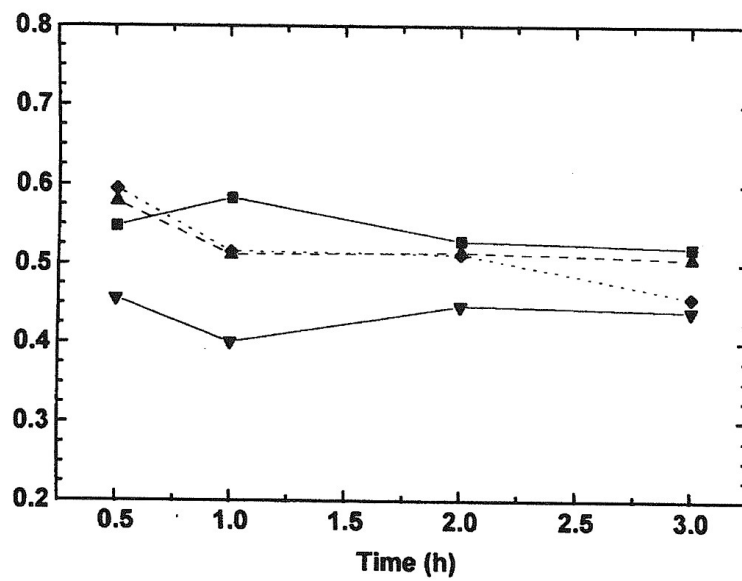


Fig. 3. Evolution in time of the variable $q_i^{A \text{ norm}}$ (on the ordinate) when no pheromone is added. Squares: $A = 0$, $D = 10$; up triangles (dashed line): $A = 10$, $D = 10$; diamonds (dotted line): $A = 20$, $D = 10$; down triangles: $A = 40$, $D = 10$.

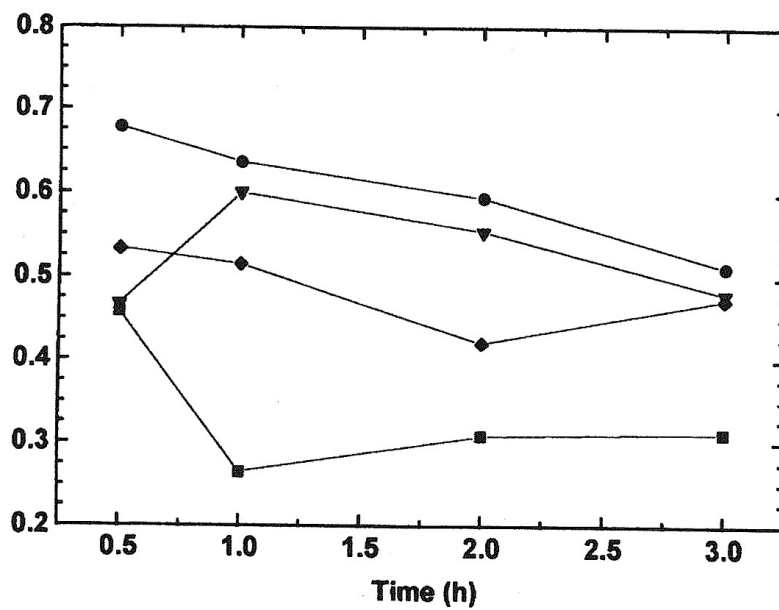


Fig. 4. Evolution in time of the variable $q_i^{A \text{ norm}}$ (on the ordinate) when pheromone is added. Squares: $A = 0$, $D = 10$; circles: $A = 5$, $D = 10$; diamonds: $A = 20$, $D = 10$; down triangles: $A = 40$, $D = 10$.

RESULTS

The experiments reported here suggest that the presence and accumulation of living or dead insects in traps, whether baited with aggregation pheromone or not, interferes with the behaviour of other free specimens of red flour beetle. This could be due to the chemical substances produced by live *T. castaneum*.

According to the experimental data, the overwhelming majority of specimens which make a choice do so within the first 3 h after the starting time. As a consequence we confined the analyses to this time limit.

Figure 1 shows the time dependence of the probability of making a choice, whether A or D, where A stands for alive and D for dead in the case in which no pheromone was added to the trap, for four starting ratios of living/dead specimens (0/10, 5/10, 10/10 and 40/10).

It seems evident that specimens prefer to make a choice when either the trap is empty or it holds 10 dead specimens. When 40 living specimens are present in the trap, a large percentage of specimens prefer to remain outside both traps. Middle-range responses correlate with middle-range starting conditions. In Fig. 2 the experimental data using the same number of insects — but with pheromone added to the traps — are shown. In this case the situation is confused. No clear trend or intrinsic difference appears among the specimens regarding the traps. After 3 h, in the most favourable conditions, only 42% of the specimens had made a choice. In the previous case this represented the lowest limit.

Figure 3 shows the time dependence of the two choices (A or D) with no pheromone added. As a general rule, no definite preference for one trap over the other appears. Nevertheless, a statistically meaningful difference does appear between A and D. Between the choice of no living specimens in the trap and 10 dead specimens in the trap, a slight preference for choice A appears after 3 h (52% to 48%).

With the choice between 40 living specimens in the trap and no dead specimens, a slight preference for choice D appears after 3 h (44% to 56%).

The result is quite different when pheromone is added (Fig. 4). With a choice between no living and 10 dead specimens in the trap, the free specimens largely prefer D, with 10 dead specimens (30% to 70%).

Otherwise, no statistically meaningful preference appears between A and D where living specimens are placed in the trap, no matter their number (5, 20 or 40). To demonstrate to what extent one choice predominates over the other one, in Figs. 5 and 6 we show the time dependence of the normalised data corrected by the weighting factors previously introduced.

Even though a clear preference of A over D (for none and 10) or of D over A (for 40 and 10) appears, due to the statistical spread of the data only a weak statistical significance can be given to these results. On the other hand, when pheromone is added to the traps, a statistically meaningful preference appears: D is preferred to A, 0 and 10. No clear preference appears with 40 and 10.

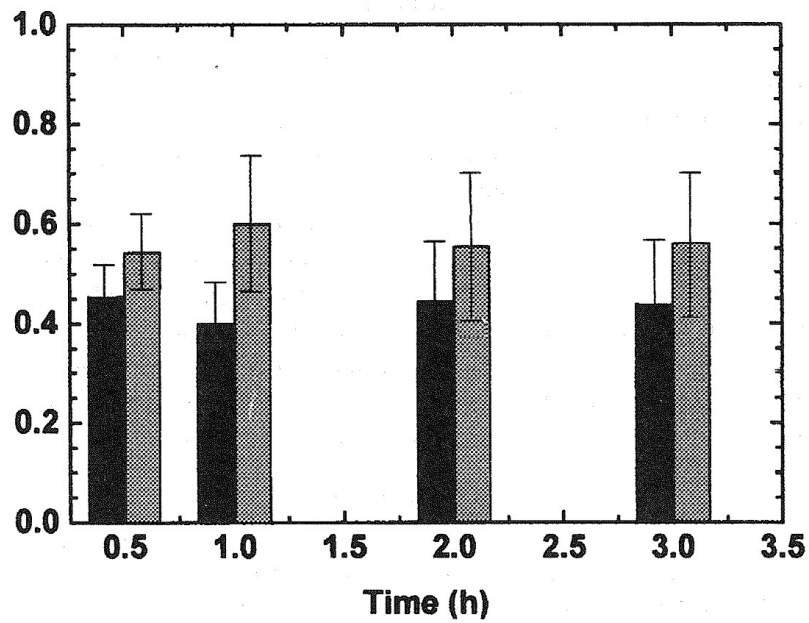


Fig. 5. Comparison of the evolution of the variables $q_i^A \text{ norm}$ (black columns) and $q_i^B \text{ norm}$ in time for the case $A = 40$, $D = 10$ without pheromone. Error bars related to each set of data are SD's.

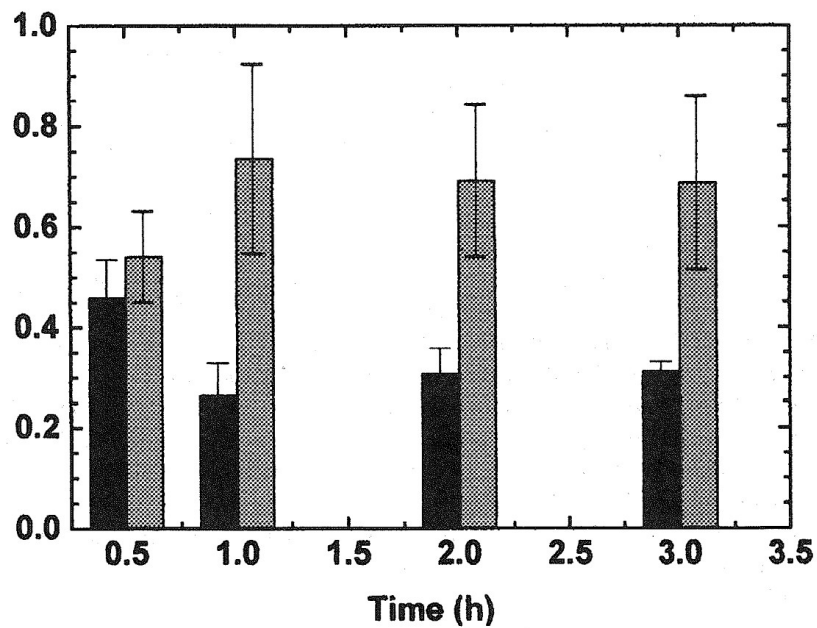


Fig. 6. Comparison of the evolution of the variables $q_i^A \text{ norm}$ (black columns) and $q_i^B \text{ norm}$ in time and for the case $A = 0$, $D = 10$ with pheromone. Error bars related to each set of data are SD's.

CONCLUSIONS

In trapping *T. castaneum*, the efficacy of traps (whether or not baited with aggregation pheromone) is influenced by the presence in them of accumulated specimens. This fact has been observed both with traps containing living insects and those with dead specimens.

The interactions of the different substances produced by *T. castaneum* adults for chemical communication are involved in this behaviour.

The results obtained in our trials suggest that living *T. castaneum* in traps attract other specimens, but under conditions of overcrowding the quinone they produce regulates population density by counteracting the effects of the aggregation pheromone, either causing the insects to disperse or suppressing aggregation behaviour.

The presence of dead insects in the traps, whether or not baited with 4,8-dimethyldecenal, can repel other free specimens of *T. castaneum*. This is probably related to the presence of a residual alarm pheromone produced by the specimens before death.

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