THE HUMIDITY RESPONSES OF *TROGODERMA GRANARIUM* EVERTS (COL., DERMESTIDAE)

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The relative humidity of the air is one of the external physical factors which may influence the damage caused by *Trogoderma granarium* Everts in granaries and breweries. According to Spangler (1965), the larvae are repelled by air of high relative humidity. Burges (1959) found that in malt stores there is a steep humidity gradient between the external layer of the stored product and its interior, but that this did not appear to influence the behaviour of larvae. He also maintains that the larvae did not seem to search for damp spots, as they occurred in higher numbers in relatively dry places in the granary.

According to Shulov (1940), the eggs and larvae of *T. granarium* require a high relative humidity for normal development. The highest concentration of beetles was found at 80–100 per cent. relative humidity. It was therefore concluded that an increase of humidity in the ambient atmosphere and the stored grain enhances the detrimental activity of the larvae.

Andrewartha & Birch (1961) write that there are some puzzling examples of insects (*Locusta migratoria* (L.), *Blatta orientalis* L.) which, when tested in a humidity gradient in the laboratory, move towards the dry end, although in nature they seem to survive and multiply better in a humid atmosphere.

The aim of the present work was to establish the response of larvae and adults of *T. granarium* to known air humidities and to find how this is influenced by different conditions.

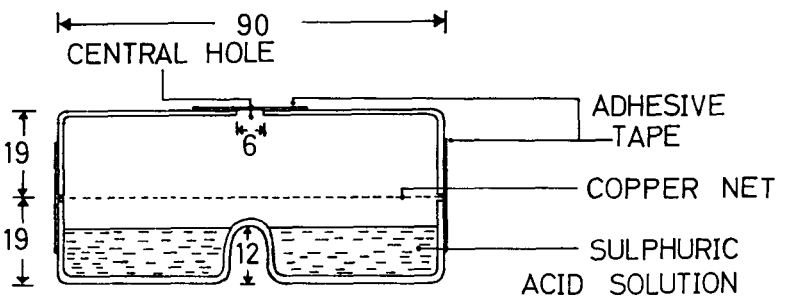

Fig. 1.—Cross-section of a humidity choice-chamber used to test the humidity responses of larvae and adults of *Trogoderma granarium* (sizes in mm.).

Experimental methods

The insects were raised on coarsely ground wheat in an incubator, at $33{\cdot}5 \pm 0{\cdot}5$°C. and 49 ± 2 per cent. R.H. The mean ages of the larvae, male adults

and female adults used in experiments were 20·4, 3·7 and 5·4 days, respectively. Specimens of uniform size were chosen.

To test the response of the larvae under various humidity gradients, we used humidity choice-chambers made of petri dishes (fig. 1). The bottom part was a dish 90 mm. in diameter and 19 mm. high, divided into two equal compartments by an impermeable partition. This dish was covered by a disc of the same diameter, made from copper netting (16 mesh/cm.), which was washed with water before every experiment and constituted an arena on which the test insects could move about. Another petri dish of the same diameter was inverted over this disc. During the experiments the bottom and the cover dishes were sealed hermetically with a wide strip of adhesive tape. At the beginning of each experiment the test insects were introduced into the choice-chamber with the aid of a funnel through a hole of 6 mm. diam. drilled in the cover dish; the hole was then sealed with adhesive tape.

Solutions of sulphuric acid of various concentrations were used to set up the required humidity gradients (Buxton & Mellanby, 1934) by placing them in the two compartments of the lower petri dish according to the relative humidity required. The solutions were replenished after every experiment. The specific gravities of the different stock solutions of H_2SO_4 were tested with a hydrometer from time to time to check that their concentrations corresponded exactly with the various relative humidities required; any deviation was corrected.

The relative humidities in the choice-chambers were determined with strips of cobalt-chloride paper, prepared according to the method of Solomon (1945). The strips were placed in the humidity choice-chambers, and their colours compared with those of other strips held in jars above standard solutions providing known and constant relative humidities. Within 15 minutes of the introduction of the solutions into the compartments one could distinguish between regions of different atmospheric humidity in the choice-chamber. Within one hour a steady state was established, ensuring a stable humidity gradient in the chamber. With solutions yielding 100 per cent. R.H. in one half of the chamber and 10 per cent. in the other, a very steep gradient was obtained at the centre of the chamber, while on either side of the chamber the relative humidities were fairly homogenous. In a combination of 30 and 70 per cent. R.H., the gradient was less steep, and for one of 50 and 60 per cent. R.H., there was a very gradual gradient of humidity across the whole chamber. Once the humidity gradient had achieved equilibrium, it remained stable for more than five days whatever the combination.

For these reasons, chambers holding the solutions were left to stay for two hours before every experiment and they were not kept for more than two days. The humidity response of the insects was therefore tested from the beginning of each experiment under conditions of a steady humidity gradient.

In variable-humidity atmospheres the temperature is known to change owing to the cooling caused by evaporation in the relatively humid region (Perttunen, 1953). In order to take this into consideration, an electronic thermometer * was employed to measure the temperatures at either side of a 20–80 per cent. R.H. gradient, the immersion-type probe of the instrument being inserted through the central hole in the lid of the chamber. Tests made in six choice-chambers under identical conditions showed an average temperature of 20·37°C. in the drier side and 20·31°C. in the moister one. These temperature differences did not appear to be significant as far as the reaction of the insects under the experimental humidity conditions was concerned.

During each experiment, the humidity choice-chambers were kept at constant temperature in a dark refrigerating incubator.

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* Model TIB, Serial G 156, Tri-R Instruments, U.S.A.

In preparing the material needed for experiments involving antennectomy of adults, the beetles were held by the abdomen with a plastic tube connected to a suction vacuum-pump ** and the antennae completely removed by lightly pressing with a pin at the scape. Special care was taken not to injure any neighbouring organs.

The readings in all the experiments were made by counting the number of beetles in each side of the arena at 15-minute intervals for two hours after the beginning of the experiment.

Results

Humidity responses of larvae and adults of T. granarium *tested separately and in groups*

In this experiment we tested the responses of larvae, males and females in groups of twenty, and also as single specimens. Counts were made eight times, at 15 min. intervals, in two hours, and were repeated on 10 different groups of insects, giving a total of 1,600 position records for each humidity gradient. The experiments with single specimens were carried out in a similar way but using 20 replications (a total of 160 position records for each gradient). The experiments were conducted in a gradient of 20–80 per cent. R.H. and at 20°C. constant temperature.

TABLE I. *The humidity responses of* T. granarium *tested in groups and singly in a gradient of 20–80 % R.H.*

			No. of records on dry side of chamber as percentage of total	Significance *
Larvae		Groups	87·25	$P < 0{\cdot}001$
		Singles	76·25	$P < 0{\cdot}001$
Adults	Males	Groups	72·94	$P < 0{\cdot}001$
		Singles	66·88	$0{\cdot}010 > P > 0{\cdot}005$
	Females	Groups	66·88	$P < 0{\cdot}001$
		Singles	61·88	$0{\cdot}100 > P > 0{\cdot}050$

* Estimated by t-test on the difference from zero of the mean excess of individuals scored in the dry side of the chamber. Values of $P > 0{\cdot}05$ regarded as not significant.

A distinct response to high humidity is indicated in Table I. In all cases, except for females tested singly, a significantly greater proportion of individuals was observed in the drier half of the chamber.

The response was greater in larvae than in adults, and in males than females, whether tested singly or in groups. The responses of all three categories were greater when tested in groups rather than singly.

TABLE II. *Humidity responses of antennectomised and normal adults of* T. granarium *in a gradient of 20–100% R.H.*

	State of antennae	No. of records on dry side of chamber as percentage of total	Significance *
Males	Intact	60·83	$0{\cdot}050 > P > 0{\cdot}025$
	Removed	45·42	$0{\cdot}400 > P > 0{\cdot}200$
Females	Intact	70·42	$P < 0{\cdot}001$
	Removed	45·42	$0{\cdot}500 > P > 0{\cdot}400$

* Estimated by t-test on the difference from zero of the mean excess of individuals scored in the dry side of the chamber. Values of $P > 0{\cdot}05$ regarded as not significant.

** Type RBF 3, made by Edwards High Vacuum Ltd., England.

The humidity response of antennectomised adults of T. granarium

We tested the responses of males and females whose antennae had been removed. They were tested singly, to avoid any group effect, and beetles with intact antennae were used as controls.

The experiment was carried out in a 20–100 per cent. R.H. gradient at 25°C., using 30 individuals of each sex, on each of which eight observations were made during two hours (giving a total of 240 position records for each sex in both treatment and control).

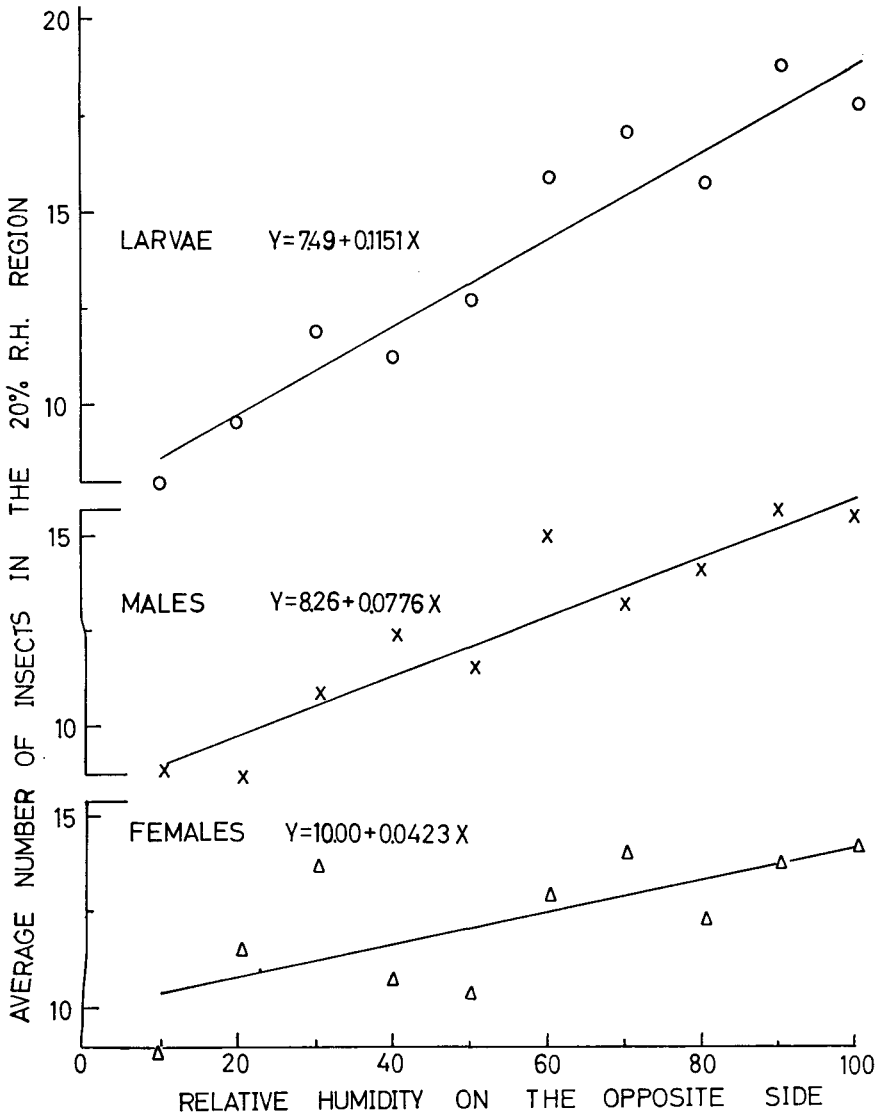

Fig. 2.—The responses of larvae and adults of *Trogoderma granarium* to different humidity gradients. Results expressed as the average number of individuals (out of groups of 20 tested) recorded on the 20% R.H. side of the choice chamber (y) for different values of the R.H. on the other side (x), with regressions of y on x.

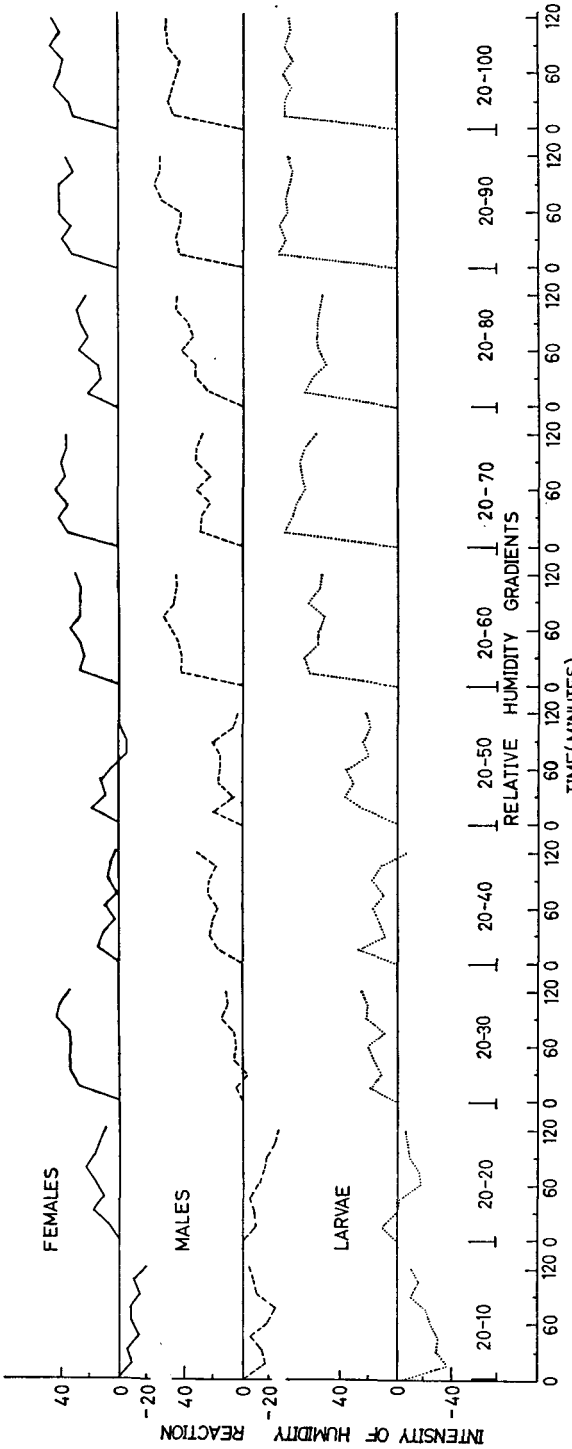

Fig. 3.—Variation with time in intensity of the reaction of larvae and adults of Trogoderma granarium to different humidity gradients.

Relative intensity $= \frac{100\ (D-W)}{D+W}$, where D is the average number of individuals at any given time on the 20% R.H. side of the chamber, and W is the number on the other side.

There was no significant response by antennectomised beetles of either sex, but the control beetles, as in the previous experiment, showed a significant preference for the drier half of the chamber (see Table II, p. 453).

Humidity responses of larvae and adults of T. granarium *in different humidity gradients*

In this experiment we tested the responses of larvae and adult beetles to ten different humidity gradients. In all cases, one of the compartments held a solution providing 20 per cent. R.H., while solutions providing a range of humidities from 10 to 100 per cent. R.H. were introduced into the other compartment. Observations on 20 individuals were made in each gradient at 15-min. intervals over two hours; each set of observations was repeated with 10 different groups of individuals, giving a total of 1,600 position records for each gradient. The temperature in this experiment was 25°C.

We calculated the average number of larvae, males and females recorded in the 20 per cent. R.H. part for each of the different humidity gradients. From these data we calculated the coefficient of linear regression of response on humidity difference for each of the three stages (larva, adult male and adult female), and plotted the respective regression lines, see fig. 2, p. 454).

A statistical analysis of the data showed that the regression coefficient differed significantly from zero ($P < 0{\cdot}001$ in larvae and males, $0{\cdot}025 > P > 0{\cdot}010$ in females).

In all cases the number of individuals recorded in the side of the chamber at 20 per cent. R.H. progressively increased as the humidity in the opposite side increased. In the same way, the average number of individuals recorded in the region at 20 per cent. R.H. was smaller than that in the other side when the latter was a lower humidity (10 per cent. R.H.). In the control experiment (20 per cent. R.H. on both sides) the average number of insects recorded on either side was approximately the same.

The degree of response after any given time in the different gradients was calculated from the formula

$$\text{Degree of response} = 100\,(D - W)/(D + W)$$

where D is the number of individuals in the 20 per cent. R.H. side of the chamber and W the number of individuals on the other side.

A positive response to 20 per cent. R.H. yields a positive value of this index.

As shown in fig. 3, the majority of the insects that respond at all to the humidity stimulus do so in the first 15 minutes, and the peak of response tends to be greater, and reached more rapidly, in the case of larvae than in that of adults, and in the steeper humidity gradients (20–60 per cent. and above) than in the gentler ones (20–50 per cent. or below). With solutions yielding 10 per cent. and 20 per cent. R.H. on either side of the chamber, a small preponderance of individuals was found at the lower humidity.

Discussion

The data of Roth & Willis (1951*b*) for other granary beetles—adults of *Tribolium confusum* Duval and *T. castaneum* Herbst—and of Willis & Roth (1950) for *T. castaneum* show a distinct negative humidity response. The response of larvae of *Tenebrio molitor* L. is also negative in various humidity gradients (Perttunen & Laherma, 1962). The results established in the present paper for *T. granarium* also show a strong negative humidity response, not unlike that found for the insects listed above. On the other hand, the results for *Sitophilus granarius* L. indicated a positive humidity response in most cases (Smereka & Hodson, 1950),

although, as with the present findings for *T. granarium*, the intensity of the response varied with the humidity gradient.

The stronger response of adults of *T. granarium* observed when they were tested in groups instead of singly is apparently due to some aggregation factor; the effect of this requires further study.

Many insects, including various stored-product beetles, have humidity receptors on their antennae (Roth & Willis, 1951*a*; Schneider, 1964). The present work proves that such organs likewise occur on the antennae of adults of both sexes of *T. granarium*.

Shulov (1955) observed normal development and propagation of *T. granarium* when exposed to 80–90 per cent. R.H., and in humid germinating grains, as well as in 0 per cent. relative humidity and grains which contain only 9 per cent. water. Lindgren, Vincent & Khrohne (1955) showed that higher humidities reduce the mortality rate of larvae and pupae of this insect at high temperatures. According to Hadaway (1956), humidities below 25 per cent. are inimical to larval development.

The results of our experiments indicate that larvae and adults of *T. granarium* show a negative humidity response in each humidity gradient tested. There is no obvious relation between this clear negative response and the humidity conditions that must exist in places where this insect develops and propagates. This question remains open and requires further study.

Summary

The humidity responses of *Trogoderma granarium* Everts were tested in humidity gradients set up in choice-chambers, each comprising a petri-dish with the lower half divided diametrically into two compartments containing sulphuric-acid solutions affording different relative humidities and surmounted by a copper-gauze platform on which the insects, released centrally, could move about. Counts of the numbers in either half of the arena were made at 15-minute intervals over two hours.

In a gradient of 20–80 per cent. relative humidity, at 20°C., the average count was significantly greater on the drier side than on the moister one for larvae and adults tested in groups and for larvae and adult males, but not females, tested singly; the response was greater when the insects were tested in groups. Antennectomised adults, tested at 25°C. in a gradient of 20–100 per cent. relative humidity, showed no response, although intact control beetles showed a significant preference for the drier side. When adults were tested in groups at 25°C. in a range of gradients, from 20–10 to 20–100 per cent., a preference for the drier side was invariably shown, the response tending to be greater and to occur more rapidly in larvae than in adults, and as the humidity gradient steepened.

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References

ANDREWARTHA, H. G. & BIRCH, L. C. (1961). The distribution and abundance of animals.—782 pp. Chicago, Ill., Univ. Chicago Pr.

BURGES, H. D. (1959). Studies on the dermestid beetle, *Trogoderma granarium* Everts. III. Ecology in malt stores.—*Ann. appl. Biol.* **47** pp. 445–462.

BUXTON, P. A. & MELLANBY, K. (1934). The measurement and control of humidity. —*Bull. ent. Res.* **25** pp. 171–175.

HADAWAY, A. B. (1956). The biology of the dermestid beetles, *Trogoderma granarium* Everts and *Trogoderma versicolor* (Creutz).—*Bull. ent. Res.* **46** pp. 781–796.

LINDGREN, D. L., VINCENT, L. E. & KHROHNE, H. E. (1955). The khapra beetle, *Trogoderma granarium* Everts.—*Hilgardia* **24** pp. 1–36.

PERTTUNEN, V. (1953). Reactions of diplopods to the relative humidity of the air. —*Annls zool. Soc. zool. bot. fenn.* **16** pp. 1–69.

PERTTUNEN, V. & LAHERMA, M. (1962). Humidity reactions of the larvae of *Tenebrio molitor* L. (Col. Tenebrionidae) and the effect of cannibalism on these reactions.—*Annls ent. fenn.* **28** pp. 71–80.

ROTH, L. M. & WILLIS, E. R. (1951*a*). Hygroreceptors in Coleoptera.—*J. exp. Zool.* **117** pp. 451–484.

ROTH, L. M. & WILLIS, E. R. (1951*b*). The effects of desiccation and starvation on the humidity behavior and water balance of *Tribolium confusum* and *Tribolium castaneum*.—*J. exp. Zool.* **118** pp. 337–361.

SCHNEIDER, D. (1964). Insect antennae.—*A. Rev. Ent.* **9** pp. 103–122.

SHULOV, A. (1940). On a new pest of grain stores in Palestine.—*Hassadeh* **20** pp. 512–514 [*In Hebrew*].

SHULOV, A. (1955). A contribution to the ecology of *Trogoderma granaria* Everts.— *Proc. Indian Acad. Sci.* (B) **42** pp. 1–13.

SMEREKA, E. P. & HODSON, A. C. (1959). Some humidity and light reactions of the granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Curculionidae). —*Can. Ent.* **91** pp. 784–797.

SOLOMON, M. E. (1945). The use of cobalt salts as indicators of humidity and moisture.—*Ann. appl. Biol.* **32** pp. 75–85.

SPANGLER, H. G. (1965). Reactions of the larvae of the khapra beetle and *Trogoderma parabile* to certain food substances and organic compounds.— *J. econ. Ent.* **58** pp. 212–218.

WILLIS, E. R. & ROTH, L. M. (1950). Humidity reactions of *Tribolium castaneum* (Herbst).—*J. exp. Zool.* **115** pp. 561–587.