Temporal Distribution of *Tribolium castaneum* Herbst and *Cadra cautella* Walker on Temperature Gradients

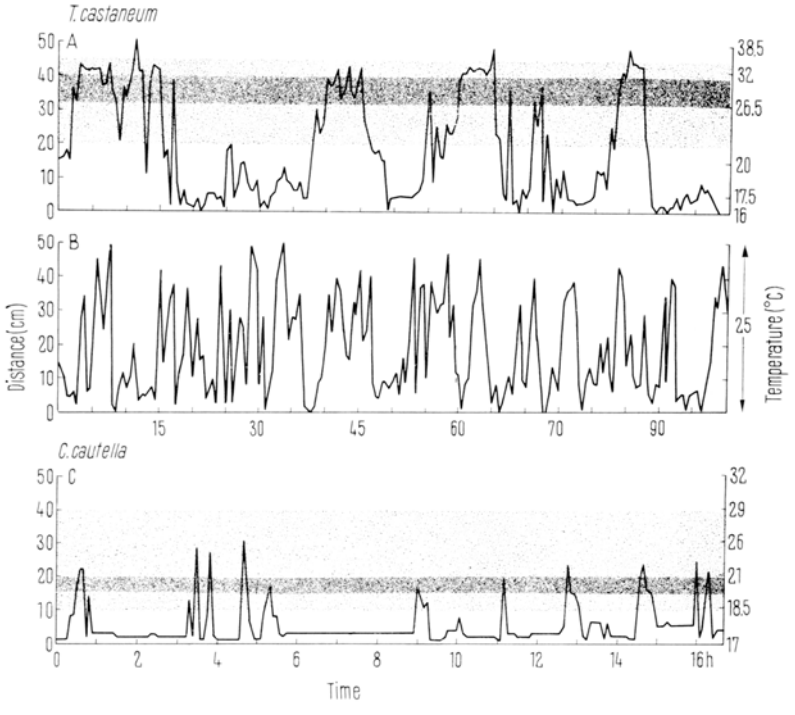
Preliminary to a study of the behaviour and biology of populations of *Tribolium castaneum* Herbst (Col., Tenebrionidae) and *Cadra cautella* Walker (Lepid., Phyticidae) on various temperature gradients, a number of experiments were carried out to examine the behaviour of individual adults.

In experiments in which 35 *T. castaneum* were examined, temperature gradients ranging from about 16 to 38 °C were used in a channel apparatus (50 cm long) containing finely-divided wheatflour as food. The concomitant inverse humidity gradient present in the apparatus was not measured. Female adults (of mixed ages) used in the tests were bred at 25 °C and 65–70% relative humidity (R.H.). The channel apparatus had a number of tracks into each of which one adult was placed. Observations on the position of each adult in its respective track were made in some tests at 5 min intervals for 4 days, and in others at 30 min intervals for 14 days and more, by means of electronic flash photography[1]. All experiments were carried out in darkness except for the intermittent period of electronic flash ($^1/_{1000}$ sec duration).

Although there was considerable variation between individuals, it was found that all adults did not remain indefinitely in one particular region of the gradient but tended to spend alternate periods in the warmer and cooler regions, with a certain degree of wandering taking place in each (Figure 1A). For example, 1 adult spent about 20 h ($17^1/_2$–$37^1/_2$ h) in the cool region of the gradient, which ranged from 16 to about 21 °C, and then about 10 h ($37^1/_2$–$47^1/_2$ h) in the warm region, which corresponded to temperatures ranging from about 21 to 32 °C. This behaviour was not observed when the same apparatus was maintained at a uniform and constant temperature of 25 °C (control experiment) (Figure 1B). The adult wandered about the apparatus with no one region being favoured more than another, with the possible exception of the ends due to 'end-effect', which is a characteristic feature of the channel apparatus[2]. This indicated that the behaviour observed in the gradient was in response to temperature (and possibly to humidity[3]) and not to any artefact induced by apparatus design.

The periodic selection of warm and cool regions in the gradient suggested that some form of cyclic behaviour was present and a periodogram analysis[4], commencing at 6 h and by steps of 1 h terminating at 48 h, for each adult was carried out. The analysis indicated that cyclic behaviour was present, though it varied from one adult to another, with a number of periodicities involved, most of which occurred between 20 and 40 h. Observations were also made on the sites selected for oviposition by adults whilst on the gradient (Figure 1A, shaded area). It was found that eggs were laid at temperatures ranging from 22–32 °C, most occurring in the 26–30 °C region, which corresponded to the warm region periodically selected by the adult. It is a matter for speculation as to whether the phase of adult behaviour which results in a selection for the warm region of the gradient is related to oviposition behaviour. Detailed examination of the position records revealed that, on the whole, the individual was more active in the warmer than in the cooler region of the gradient. This difference in activity, measured by the extent by which the individual had moved between successive records in these 2 regions, is believed to reflect the general effect of temperature on the overall activity of the adult.

[1] T. G. AMOS, Anim. Behav. *13*, 558 (1965).
[2] D. L. GUNN and J. S. KENNEDY, J. exp. Biol. *13*, 450 (1936).
[3] T. G. AMOS and F. L. WATERHOUSE, Oikos *18*, in press (1967).
[4] E. T. WHITTAKER and G. ROBINSON, *The Calculus of Observations* (Blackie, London 1924).



(A) Representative data for a female adult of *Tribolium castaneum* on a temperature gradient maintained in a channel apparatus. The shaded area denotes the region in which eggs were laid by females whilst on the gradient, with the densely shaded area indicating the region in which most eggs were laid. (B) Representative data for a female adult of *T. castaneum* in the same channel apparatus maintained at a uniform and constant temperature. (C) Representative data for a female adult of *Cadra cautella* on a temperature gradient maintained in a channel apparatus. The shaded area denotes the region in which eggs were laid by females whilst on the gradient, with the densely shaded area indicating the region in which most eggs were laid.

In similar experiments in which over 30 *C. cautella* were examined, temperature gradients ranging from about 17–32 °C were used without food present in the channel apparatus. Adults (females) were 1–3 days old and had been bred at 25 °C and 65–70% R.H. Observations were taken at 5 min intervals for about 6 days. Representative data is given in Figure 1C where it can be seen that the female adult spent alternate periods in the cool region of the gradient interspersed with periods, of a much shorter duration, in a warmer region. Oviposition occurred in the warm region of the gradient where the adult was also relatively more active.

Although the experiments reported above can only be considered as exploratory, it is interesting to note that GRAHAM[5] inferred from his population gradient studies that some form of cyclic behaviour was present in *T. castaneum*. Cyclic behaviour has also been observed in *Pseudophonus pubescens* for example, where daily and seasonal periodicities were present[6]. There appears to be no evidence in the literature suggesting cyclic behaviour of *C. cautella* on a temperature gradient. Further work is planned to investigate the nature of this cyclic behaviour and it is also proposed to examine other stages in the life cycle, such as the larval stage, to determine if a similar phenomenon exists[7].

Résumé. Des observations faites en laboratoire sur le comportement d'individus adultes de *Tribolium castaneum* et *Cadra cautella* dans des zones de températures différentes montrent que ces insectes ne se trouvent pas indéfiniment dans la même zone, mais ont la tendance de séjourner alternativement dans une zone chaude et une zone froide. Ces déplacements semblent avoir un caractère rythmique.

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Department of Natural History, The University of Dundee, Dundee (Scotland), 3 August 1967.

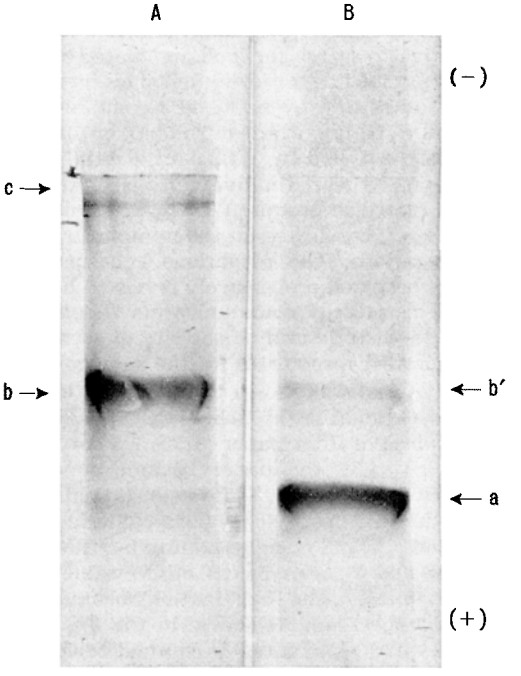
[5] W. M. GRAHAM, Anim. Behav. *6*, 231 (1958).
[6] A. V. LIKVENTOV, Ént. Obozr. *30*, 208 (1949).
[7] This work forms part of the research programme of the Department of Natural History and was carried out in collaboration with the Tropical Stored Products Centre, Ministry of Overseas Development, Slough.
[8] On transfer from Tropical Stored Products Centre, Ministry of Overseas Development, Slough, Bucks.

Electrophoretic Mobilities of Malate Dehydrogenases from Barley Seedlings

We have recently reported[1] the occurrence in young barley seedlings of 2 malate dehydrogenases (L-malate: NAD oxidoreductase, 1.1.1.37), one being associated with the cytoplasmic fraction and the other a particulate, presumably mitochondrial, fraction of the cell. The 2 enzymes have been shown to differ in chromatographic behaviour on diethylaminoethyl (DEAE) cellulose and in their substrate affinities[1]. Similar correlation between intracellular origin and chromatographic behaviour of malate dehydrogenases has also been reported for other plant tissues[2]. In the present study, the 2 enzymes have been compared as regards their electrophoretic mobility with a view to further characterizing their physical properties. Electrophoresis of the 2 subcellular fractions of barley seedlings on polyacrylamide gel, followed by detection of the enzyme activity by tetrazolium staining, revealed that the 2 malate dehydrogenases are electrophoretically heterogeneous.

The cytoplasmic and mitochondrial fractions of young barley (*Hordeum sativum*) seedlings were prepared as previously described[1]. Electrophoresis[3] of the fractions on polyacrylamide gel was carried out at 4 °C. Aliquots of samples containing 20–200 μg protein, determined by the standard KJELDAHL procedure[4] after precipitation with trichloroacetic acid, were each layered on top of the spacer gel prior to electrophoresis. Good resolutions of the enzymes were obtained by employing a current of 2 mA per column for 15 min and then 5 mA for 30 min in *Tris*-glycine buffer, pH 8.6. Malate dehydrogenase activity was detected by the tetrazolium technique[5]. The reaction mixture contained 1 vol 1.0 *M* sodium malate; 1 vol 2 mg/ml phenazine methosulphate; 1 vol 5 mg/ml nitro blue tetrazolium; 1 vol 0.1 *M* sodium cyanide; 4 vol 0.75 mg/ml nicotinamide adenine dinucleotide; and 2 vol 0.1 *M* phosphate buffer, pH 6.0.

It is clear from the Figure that the cytoplasmic and mitochondrial malate dehydrogenases, the activity of which being detected by the purple formazan bands a and



Electrophoresis patterns of malate dehydrogenases from barley seedlings. (A) Mitochondrial fraction. (B) Cytoplasmic fraction. These gels were simultaneously subjected to electrophoresis and incubated in the reaction mixture for the same period of time.