

replicable feature of their procedures. It has been suggested to me that perhaps individual animals were forced, by jabbing, pushing and so on, to initiate a trial and traverse the maze. But such inelegant procedures, while often standard practice with invertebrates, are themselves objectionable, for they may introduce systematic bias by permitting the experimenter unintentionally to guide an animal's movements². Such a bias could easily account for the small effect reported.

I thank the Department of Zoology, University of Edinburgh, and Dr A. Manning for the use of facilities while preparing this manuscript.

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Received March 19; revised June 15, 1970.

¹ Borsellino, A., Pierantoni, R., and Schieti-Cavazza, B., *Nature*, **225**, 963 (1970).

² Ratner, S. C., *Psychol. Rec.*, **14**, 31 (1964).

Reply to Comment on Survival of Learning acquired at the Larval Stage

REYNIERSE'S criticism¹ of our work on the survival of learning acquired at the larval stage raises doubts about whether learning does occur at all in larval animals.

Disregarding the long-standing discussion about the exact nature of learning (for example, ref. 3), our criterion for maze learning should satisfy the requirements mentioned by Reynierse. Considering only the results of the first trial of each day, we take for the k^{th} day a statistical variable x_k , with values ± 1 ; $+1$ for a correct choice, -1 for a wrong choice. We plot the total displacement

$$X_N = \sum_{k=1}^N x_k$$

due to the accumulation of ± 1 steps until N^{th} day. A few typical examples of such runs are shown in Fig. 1. These data show that after an initial period of chance responses, there are increasingly long runs of correct choices.

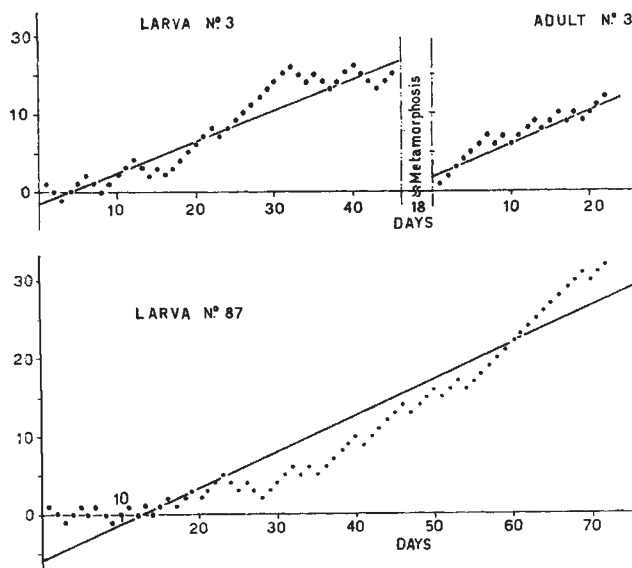


Fig. 1. Two examples of runs. The upper one shows also the adult performance after metamorphosis. The ordinate shows the cumulative sum of daily steps $+1$ (correct choice), or -1 (wrong). The slope of the fitted straight line is a measure of the general trend.

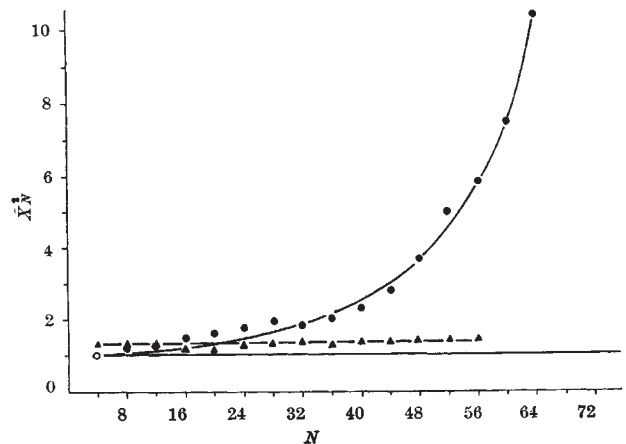


Fig. 2. Ratio of the variance $\overline{X_N^2}$, taken over the population, to the expected value N for random behaviour. See also text. ●, Learning group; ▲, control group.

We fitted the 200 individual diagrams with straight lines—using the least square criterion—to obtain a parameter measuring their trends (Fig. 1). While the distribution of the slopes of such lines for the control group is symmetrical around slope zero, within statistical fluctuation (positive/negative = 48/44), the distribution for the trained group is definitely asymmetrical (positive/negative = 62/38).

The cumulative variable X_N , when averaged over the population for random choices, gives $\overline{X_N} = 0$; its average square instead has $\overline{X_N^2} = N$. So the ratio $\overline{X_N^2}/N$ ought to be constant and equal to 1 for a population of individuals making completely random choices. Although this ratio is constant (and equals 1.2) for the control group, for the trained larvae it increases regularly with time (Fig. 2). The performance of the trained population improved continuously.

We believe that we have answered the questions raised about the ability of larvae of *Tenebrio molitor* learning in a maze. As for their motivations in so doing, we have nothing new to add to the "explanations" or hypotheses that are already well known² and used of the many theories advanced so far.

We were very careful in planning and in executing our experiments, which were started in September 1964, repeated in better controlled conditions in 1966 and reported as a preliminary note in 1967⁴. We believe that these results indicate that the animals exhibited learning behaviour rather than the experimenters.

After we had written this reply, we received a reprint⁵ which described the training of *Tenebrio molitor* larvae followed by testing for the persistence of memory traces after metamorphosis. Apart from small differences in detail, the conclusions were the same as ours.

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Received April 7, 1970.

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⁴ Borsellino, A., Pierantoni, R., and Schieti-Cavazza, B., *Atti Cong. Gruppo Naz. di Cibernetica*, Pisa, 20-21, 29 (1967).

⁵ Cherkarshin, A. N., Shermain, I. M., and Stajekhina, V. S., *Akad. Nauk. USSR Physiol.*, special issue, 117 (1968).