

# THE TEMPERATURE PREFERENCES OF THE MOTILE STAGES OF *STOMOXYS CALCITRANS* LINNAEUS (DIPTERA: MUSCIDAE)

B. SUTHERLAND, Veterinary Research Institute, Onderstepoort 0110

### ABSTRACT

SUTHERLAND, B., 1980. The temperature preferences of the motile stages of *Stomoxys calcitrans* Linnaeus (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research*, 47, 7-11 (1980).

When adult *Stomoxys calcitrans* were exposed to a temperature gradient, 66% of them selected temperatures between 20,1 and 32,5 °C. The larval stages preferred temperatures between 19,5 and 33,2 °C. Although the differences in the temperature preferences of the different larval stages were not significant, the fully-fed larvae appear to prefer slightly cooler conditions than the feeding stages. The temperature preferences of both the adults and the larvae are not influenced by the temperatures to which the developmental stages of the experimental flies or their previous generations were exposed.

### Résumé

LES PRÉFÉRÈNCES DE TEMPÉRATURE DES STADES MOBILES DU *STOMOXYS CALCITRANS* LINNAEUS (DIPTERA: MUSCIDAE)

Quand les adultes de *Stomoxys calcitrans* furent exposés à une gradient de température, 66% d'entr'eux choisirent des températures entre 20,1 et 32,5° C. Les stades larvaires préférèrent des températures entre 19,5 et 32,2° C. Bien que les différences dans les préférences de température des différents stades larvaires ne furent pas significatives, les larves entièrement alimentées semblent préférer des conditions légèrement plus froides qu'aux stade d'alimentation. Les préférences de température des adultes et des larves ne sont pas influencées par des températures auxquelles les stades de développement des mouches expérimentales ou de leurs générations antérieures furent exposées.

### INTRODUCTION

Nieschulz (1934), who studied the temperature preferences of *Stomoxys calcitrans* adults by exposing them in a temperature choice chamber, found that after exposure periods of 24 h or more their preferences were affected by external factors, such as food and humidity, and he therefore distinguished between short- and long-term temperature preferences.

Deal (1941) defined temperature preference as 'the temperature to which an insect moves if given its choice of a temperature gradient'. However, the preferred temperature is not a definite point on the scale but rather, because of individual variability, a temperature zone (Deal, 1941; Belehradek, 1957). Precht, Christophersen, Hensel & Larcher (1973) have therefore suggested that the temperature preferences of insects be expressed as mean temperature values, plus or minus their standard deviations (S.D.), and that these temperature zones should include 66% of the total number of observations.

In this study a temperature choice chamber was used to determine the temperature preferences of the various motile stages of *S. calcitrans*.

### MATERIALS AND METHODS

#### Temperature choice chamber (Fig. 1)

The temperature choice chamber was double-walled, with aluminium on the inside and galvanized iron outside; the space between these 2 walls was insulated with polystyrene. The inner box was 1 000 mm long, 50 mm wide and 80 mm deep. Its base was a solid aluminium bar, with a U-shaped tube across it at each end. The temperature gradient was created by circulating hot water (50 °C) with a Lauda Thermo-Temp water pump through the U-shaped tube at one end of the base and circulating ethylene glycol (0 °C) with a Paratherm II electronic water-bath through the other end. The lid was made of transparent perspex and along its axis it had 10 small holes, each containing a mercury thermometer, plus 3 larger holes fitted with rubber stoppers, one near each end

and one in the centre. A 20 Watt cool white neon light, 600 mm long, was held in position by means of a retort-stand, 1 000 mm above the chamber.

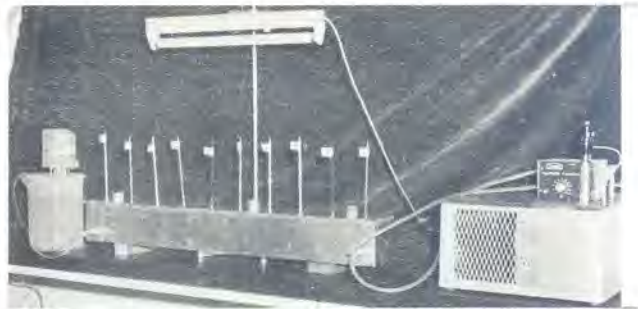


FIG. 1 Temperature choice chamber with heating and cooling pumps and neon light in position

The temperature choice chamber was used in a temperature controlled room (22 °C). The temperature gradient in the chamber was calibrated with a Rotronic micro-climate recorder. In the 'clean-dry' chamber there was a small vertical temperature gradient of 0,5-2,0 °C and a mean temperature gradient for this 'clean-dry' chamber was calculated from several temperature recordings (Table 1). There also was a horizontal humidity gradient in the chamber from 26% at the heated to 40% at the cooled end.

TABLE 1 Temperature gradient in temperature choice chamber

'Clean-dry' (°C)	Filled with larval breeding medium (°C)
37,3	43,0
33,3	39,2
31,1	33,8
28,8	31,0
26,3	27,5
23,6	23,0
21,6	21,1
18,7	17,0
16,3	12,8
14,0	9,3

*Adult temperature preference studies*

The short-term temperature preferences of adult *S. calcitrans* were determined in the 'clean-dry' temperature choice chamber, with the neon light as additional light source. Flies 18–24 hours old, 3–4 days old, and 8–10 days old from the stock colony were used (Sutherland, 1978). Three–4-day-old flies from the stock colony, maintained from eclosion until exposure at 20 °C, were also used to determine whether their thermal history had any effect on their temperature preferences.

Flies of the required age group were collected from the stock colony and lightly anaesthetized with CO<sub>2</sub>. Twenty flies of the required sex were then selected and placed in a test-tube plugged with a damp cotton-wool stopper; the remaining anaesthetized flies were discarded. The test-tube containing the flies was placed in an incubator at either 27 °C or 20 °C, depending on their previous temperature exposure, for 30 minutes before being released in more or less equal numbers through the 3 large openings in the lid of the pre-treated chamber.

Observations commenced after 15 minutes and the number of flies in each section of the chamber was counted every 10 minutes for the following hour. The temperatures registered by the 10 thermometers were recorded before each examination. At the end of each trial the flies in the chamber were killed with diethyl ether. Thereafter the chamber was thoroughly washed with water and allowed to dry.

*Larval temperature preference studies*

These experiments were conducted in daylight without any extra light source. The chamber was lined with aluminium foil and a 3–4 mm layer of 8-day-old fermented larval breeding medium (lucerne 800 g, wheat bran 200 g, water 2 000 ml), was added to it. Prior to use, this breeding medium was evenly moistened with water and thoroughly mixed to prevent the accumulation of any free water in the chamber. There was no vertical temperature gradient in this medium, nor did it dry out during a 4-day period while exposed to the temperature gradient in the choice chamber.

From 80–100 of the youngest larvae of each required developmental stage were seeded over the entire length of the chamber. Newly-hatched larvae, obtained from batches of eggs from the stock colony that had been transferred to moist filter paper strips in Petri dishes kept at 27 °C, were transferred to the choice chamber with a soft camel-hair brush. The other larval stages were obtained by gently washing the contents of the larval breeding jars through brass sieves, of which the finest had a mesh aperture of less than 1,0 mm. The ends of the aluminium foil lining were then folded over the breeding medium in such a way that a strip of medium was exposed down the length of the chamber. When the lid was placed in position the bulbs of the thermometers were buried in the strip of exposed medium.

The larvae were left for 1 h to orientate themselves in the chamber before the heating and cooling pumps were switched on. The 3 instars of feeding larvae were exposed simultaneously for 24 h, while the fully-fed 3rd instars were exposed separately for a 72 h period. In some experiments larvae from the stock colony were used; in others, designed to determine whether the

thermal histories of the larvae have any effect on their temperature preferences, both feeding and fully-fed 3rd instar larvae raised for 3 generations at a culturing temperature of 30 °C, but otherwise under the same experimental conditions as those from the stock colony, were also used.

At the end of each exposure period the lid and thermometers of the choice chamber were removed rapidly and the breeding medium was immediately partitioned off into sections between the thermometers. Subsequently, the medium plus the larvae from each section in the chamber was transferred to a separate 500 ml non-waxed paper cup.

Within an hour of its removal from the chamber, each sample of breeding medium was examined under a table model magnifier. In the case of the feeding stages all the live larvae were transferred to jars containing Carnoy's fluid (absolute ethanol 60 ml + chloroform 30 ml + glacial acetic acid 10 ml) and later examined under a stereoscopic microscope to determine the numbers collected from the different sections of the temperature gradient. When 3rd instar larvae were left for 72 h in the choice chamber, the pupae in each section were counted directly.

*Statistical analyses*

All experiments on the temperature preferences of the various motile stages of *S. calcitrans* were repeated 3 times. The data were statistically analysed and histograms, reflecting the mean  $\pm$  S.D. (standard deviation) of the temperature preferences of 66% of the test insects from each of the various treatments, were constructed.

## RESULTS

*Temperature preferences of adults*

The temperature preferences of the adults are illustrated histographically in Fig. 2.

The mean preferred temperatures of the various groups of adults from the stock colony were:

18–24-hour-old males: 25,2 °C  $\pm$  5,1.

18–24-hour-old females: 25,5 °C  $\pm$  4,0.

3–4-day-old males: 28,9 °C  $\pm$  3,6.

3–4-day-old females: 26,4 °C  $\pm$  4,6.

8–10-day-old males: 25,5 °C  $\pm$  3,8.

8–10-day-old females: 26,4 °C  $\pm$  4,0.

The mean preferred temperatures of 3–4-day-old adults from the stock colony, maintained since eclosion at 20 °C, were:

3–4-day-old males: 27,0 °C  $\pm$  4,6.

3–4-day-old females: 25,8 °C  $\pm$  4,7.

There were no significant differences between the temperatures preferred by the various groups of adult flies.

*Temperature preferences of the larvae*

The temperature preferences of the larvae are illustrated histographically in Fig. 3.

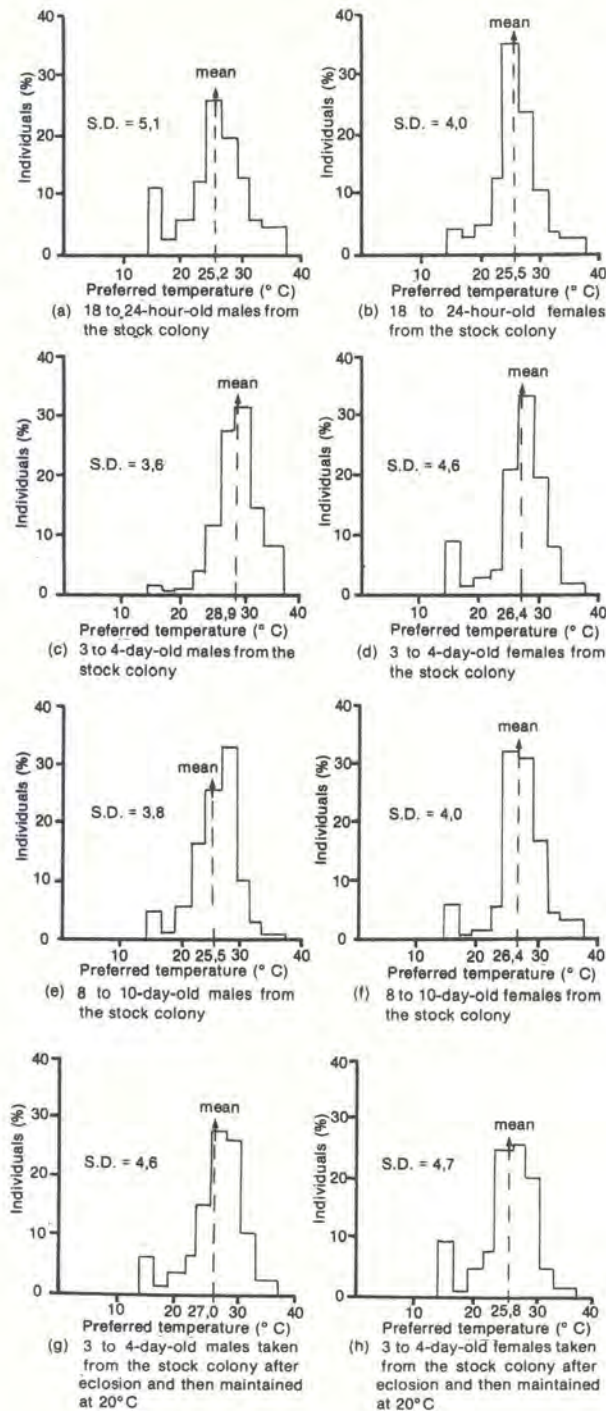


FIG. 2 The mean temperature preferences of *Stomoxys calcitrans* adults

The mean temperature preferences of larvae collected from the stock colony were:

- 1st instar larvae:  $28.2^{\circ}\text{C} \pm 4.6$ .
- 2nd instar larvae:  $28.7^{\circ}\text{C} \pm 4.5$ .
- 3rd instar feeding stage:  $26.5^{\circ}\text{C} \pm 5.2$ .
- Fully-fed 3rd instar larvae:  $24.3^{\circ}\text{C} \pm 3.6$ .

The mean temperature preferences of 3rd instar larvae raised at  $30^{\circ}\text{C}$  were:

- 3rd instar feeding stage:  $26.3^{\circ}\text{C} \pm 5.0$ .
- Fully-fed 3rd instar larvae:  $23.4^{\circ}\text{C} \pm 3.9$ .

There were no significant differences in the temperature preferences of the various larval stages.

## DISCUSSION

*S. calcitrans* is a eurythermic species and must therefore adapt to diurnal and seasonal temperature changes. The variations in temperatures to which the flies were exposed during their development did not affect their temperature preferences, which appear to be inherent.

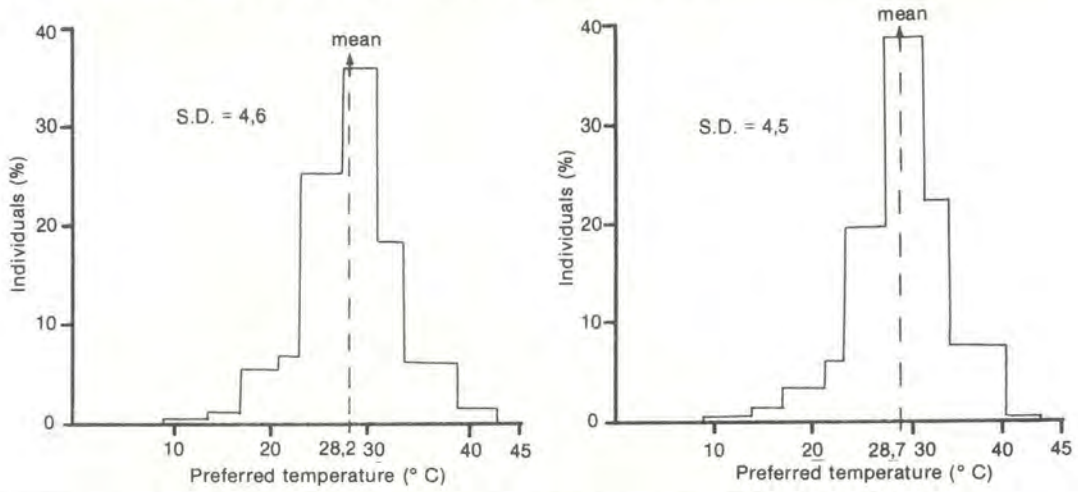
Adults show definite temperature preferences which are not influenced by their age or sex, but they are unable to discriminate between small temperature differences. Their ability to avoid unsuitably high or low temperatures results in the selection of optimum conditions for breeding sites and thus contributes to their success as a species. Nieschulz (1934) found that in the short-term the temperature preferred by *S. calcitrans* was  $28.7^{\circ}\text{C}$ , irrespective of their age, sex, culturing temperature or culturing relative humidity. He concluded, however, that 'the preferred temperature is a property of the species which is not dependent upon climatic influences'. Spencer, Pitts & Bay (1976) also found that neither the humidity history nor the sex of the stable fly had any effect on its temperature preferences, while Jack & Williams (1937) found that light had no effect on the latter phenomenon. Differences in adult temperature preferences in this study and those of Nieschulz (1934) may result from the different analytic methods that were used.

Thomsen & Thomsen (1937) found that the feeding and non-feeding larval stages of the stable fly preferred the same temperatures, a fact which was statistically proved in this study. These authors found that 59.3% of their experimental larvae selected temperatures between  $22.8$  and  $29.8^{\circ}\text{C}$ , while Hafez & Gamal-Eddin (1961) found that the selected temperatures were between  $15$  and  $30^{\circ}\text{C}$  for the feeding stages and between  $15$  and  $25^{\circ}\text{C}$  for the fully-fed stages. The latter authors' results suggest that, although there were no significant differences between the temperature preferences of the different stages, the older larvae, especially the fully-fed stage, preferred lower temperatures (Fig. 3). This preference would account for the fact that pupation always occurs towards the periphery of the breeding site, where temperatures are normally lower than in the interior (Sutherland, unpublished data, 1978). This temperature-induced selection of pupation sites may also contribute to the success of the species in its natural habitat in that this site must allow newly-emerged flies to reach the outside air without being trapped in the immature compost-like habitat after emergence.

## ACKNOWLEDGEMENTS

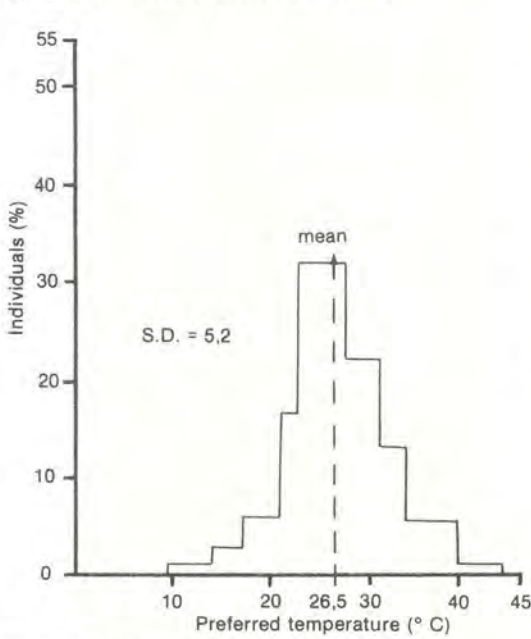
The author is indebted to Prof. H. E. Paterson, University of the Witwatersrand, Dr I. G. Horak, University of Pretoria, and Dr C. J. Howell, Onderstepoort, for their comments on the manuscript. Sincere thanks are also due to: Dr Anna Verster for editing the manuscript, Prof. K. R. Solomon, University of Guelph, for helping with the statistical analyses, Mr J. S. Marais, Onderstepoort, for constructing the temperature choice chamber, Mrs Cynthia Liebenberg for drawing the histograms and Mr A. M. du Bruyn for taking the photographs.

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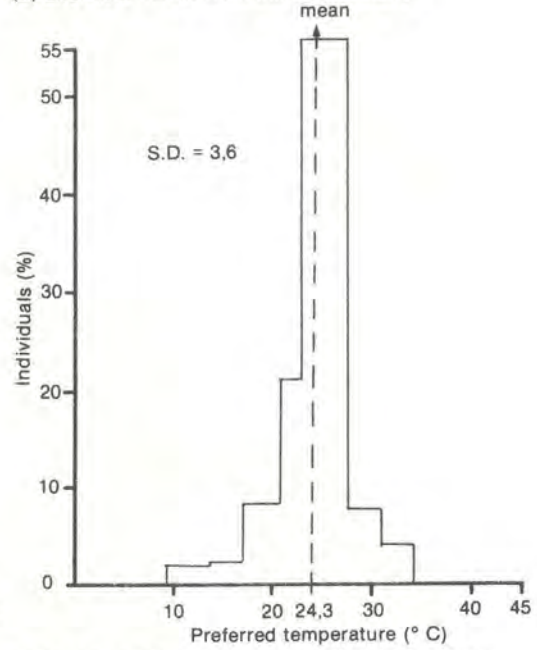


(a) 1st instar larvae from the stock colony

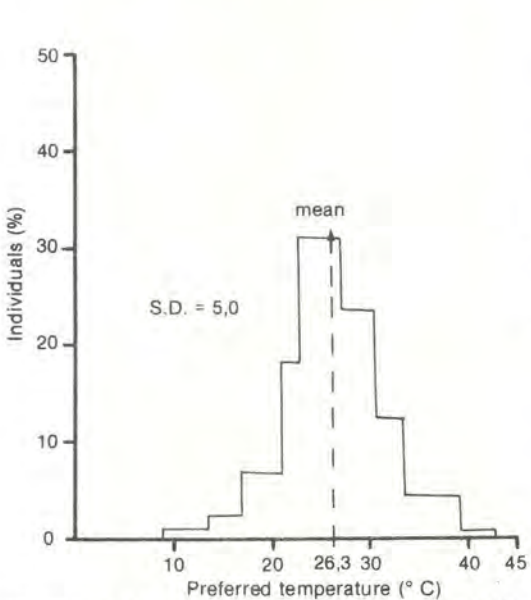
(b) 2nd instar larvae from the stock colony



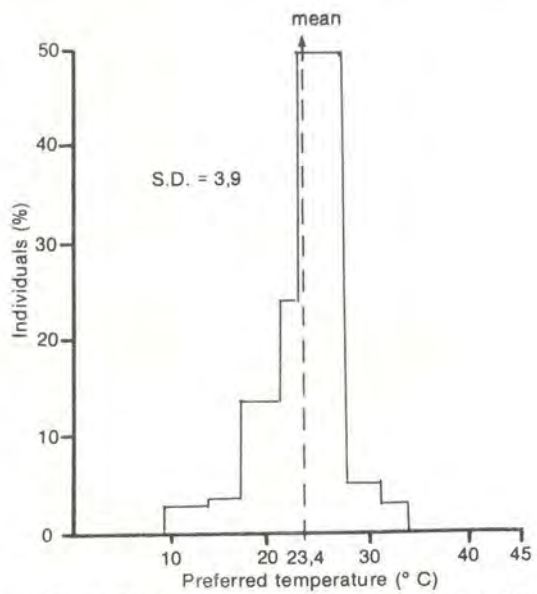
(c) 3rd instar feeding larvae from the stock colony



(d) 3rd instar fully-fed larvae from the stock colony



(e) 3rd instar feeding larvae from the sub-colony maintained at 30° C



(f) 3rd instar fully-fed larvae from the sub-colony maintained at 30° C

FIG. 3 The mean temperature preferences of *Stomoxys calcitrans* larvae

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