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Warm and cold receptors of two sensilla on the foreleg tarsi of the tropical bont tick *Amblyomma variegatum*

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Summary. A pair of antagonistic thermal receptors has been identified in each of two long, tapering, poreless setae located distally on the foreleg tarsi of the tropical *bont* tick, *Amblyomma variegatum* (Fig. 1). One, the cold receptor, responds to a rapid *drop* in temperature (T) with a sudden rise in impulse frequency (F). The other, a warm receptor, responds to a rapid *rise* in T with a sudden rise in F (Figs. 2, 4). These two units are unusual for sharing their seta with two other units which are mechanosensitive. The four are distinguishable on the basis of spike amplitude and form (Fig. 3). Hence the thermal sensitivity displayed is hardly attributable to the pair of cells with tubular bodies but rather to the two extending up into the seta (for structure, see Hess and Vlimant 1982, 1983a).

As based on the first 100 ms of the response, differential sensitivity to rapid T change is -16.1 ± 10.4 (imp/s)/°C for cold units, 17.6 ± 9.5 (imp/s)/°C for warm (Table 1). As progressively larger segments of the spike train are employed to determine F, differential sensitivity of the warm unit drops off much more quickly than that of the cold (Table 2, Figs. 5, 6). In the cold unit resolving power (the difference in rapid temperature change discriminable with 90% probability by a pair of responses of a single unit at average sensitivity) continues to increase as the segment of the spike train determining F is lengthened (from 0.58 °C for 100 ms segments to 0.41 °C for 1,100 ms segments). Resolving power of the warm unit, on the other hand, tends to decrease as longer segments are employed (from 0.52 °C for the first 100 ms to 0.80 °C for the first 1,100 ms). These relationships provoke the question of whether the spike trains may be evaluated in the CNS in different fashions.

Abbreviations: b slope of characteristic curve; F impulse frequency in impulses per second (imp/s); n number of individuals examined; Pw partial pressure of water vapor in Torr; r correlation coefficient; s SD of responses from characteristic curve; SD standard deviation; T temperature in °C; ΔT difference in T. Refers to difference between initial and end temperature in abrupt T changes

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Introduction

When the tick, *Ixodes ricinus*, is placed in a radial temperature organ, it tends to remain in an area within 3–5 °C of its preadaptation temperature (Totze 1933). According to Lees (1948) the same species approaches a sharp temperature boundary much closer when its forelegs are amputated than when it is intact. Though the operation is traumatic, the latter experiment strongly suggests that there are thermoreceptors on these appendages. The sensory unit found by Wallade et al. (1981) in a seta on the front tarsi of *Rhipicephalus appendiculatus* strengthens this suspicion. Impulse frequency of the unit rose by a factor of 10 when a stream of air 3–4 °C cooler than room temperature was trained on it. It fell silent when suddenly confronted by a stream of warm air, as it also did when an incandescent light was focused on it. When the light was turned off, its activity increased by an amount which appeared to depend on the prior intensity of the light. Though lateral pressure from the air stream may distort the seta slightly, the unit may well be responding in each case to the change in temperature as such. A priori it is also conceivable that the unit is primarily a hygroreceptor and that the increased firing rate upon cooling is in reality a response to a temperature-induced change in relative humidity. These hypotheses need testing.

Except for the above indications, almost nothing was known of the physiology of tick thermal receptors prior to the present investigation. Pilot examination leading up to three recent papers on front tarsal sensillum structure in the tropical *bont* tick, *Amblyomma variegatum* (Hess and Vlimant 1982, 1983a, b) had revealed several types of poreless sensillum there, one of which seemed an interesting candidate for physiological study. Though not the same sensillum as Wallade recorded from, Type np/C was long, easily accessible and somewhat isolated. It is innervated by two cells with tubular bodies and by two others extending well up into the hair shaft. The absence of wall pores suggested none was chemosensitive. If any was a thermoreceptor and demonstrably not mechanosensitive, the combination of modalities in a single sensillum would be unusual, at least as compared with those found so far in insects (cf. review: Altner and Prillinger 1980).

As things turned out, there are two antagonistic thermoreceptors in np/C sensilla responding with an increase in impulse frequency, one to rapid drops in temperature (the cold unit) and the other to rapid rises (the warm unit). Not infrequently both were encountered in a single sensillum. In contrast to the warm unit found on the antennae of the larvae of the cave beetle, *Speophyes lucidulus* (Loftus and Corbière-Tichané 1981), this one was easier to contact. As a result the quantity of data assembled was sufficient to permit a conservative estimate of its resolving power for rapid temperature changes.

Since additional thermoreceptors probably belong to *Amblyomma*'s complement, the investigation begun here must be pressed further before the contribution can be determined which thermoreceptors make towards the survival of this important 3-host tick between feedings and towards finding its prey. To control it would be very desirable. For not only is it a vector for Q-fever in man and animals and also for heartwater disease in farm animals throughout much of Africa. The tick has also been reported in the West Indies and Guatemala (Hoogstraal 1956).

Material and methods

The *Amblyomma variegatum* adults used in these experiments were reared on bovines (Domaine des Barges, Vouvry, Switzerland) and generously placed at our disposal by Ciba-Geigy Ltd. After the second and final moult they were kept unfed in a plexiglass container for a period of 2–6 months at 25 °C and 92% rel. humidity, set by a concentrated $Na_2CO_3 \cdot 10H_2O$ solution. For ticks even longer periods of fastening are the rule (Balashow 1972).

Preparation and recording. A foreleg of an intact tick was positioned in a shallow furrow along the edge of a narrow plexiglass prism and attached with small strips of Scotch Tape. Illumination of the tarsus by either transmitted or reflected light was therefore possible; a combination of the two was usually employed. After mounting, the coxa was partially crushed so as to eliminate muscle spiking by traumatizing the efferent nerve, without interrupting the flow of hemolymph. Such preparations held many hours.

Impulses were recorded between electrolytically sharpened tungsten needles, one inserted into the cuticle of the sensillum base at an angle of about 15° with respect to the seta, and the other into the tarsal scissure just proximal to the sensillum. Amplification procedures were standard.

Stimulation was provided by three air streams constantly emerging at 2.5 m/s from jets 7 mm in diameter and 20 mm from the preparation. Two of the streams (A and B) were at the same temperature but at different partial pressures of water vapor. Two others (B and C) were at different temperatures but the same partial pressure of water vapor. One or other of the streams was playing at all times on the preparation.

The rapid-change stimulation employed in these experiments consisted in switching from one stream to another for 1.3 s and then back to the first. Switching was done by electromagnets. When a second jet was snapped into position, an attached gate diverted the first stream but did not interrupt it. Since all three streams were at terminal values of temperature (T) and partial pressure of water vapor (Pw) before a switch occurred, the transition from one set of stimulus conditions to a second set involved little more than the 12–15 ms needed for one air stream to replace another.

A 3-min recovery period in the first air stream was permitted after each rapid-change stimulation. As indicated by a sample of 326 such periods, T displayed little drift during them: 0.08 ± 0.10 °C (mean and standard deviation). The average rate of T change independently of its direction for such periods was usually below 0.001 °C/s and almost never reached 0.002 °C/s. Further, the total drift in T even during protracted series of stimulations rarely exceeded 0.5 °C. For the purposes of this investigation neither the speed nor the extent of the drift appeared sufficient to demand further attention.

During recovery periods T or Pw of the second stream could be altered in preparation for the following rapid change. Changes undertaken in initial T or Pw of the first stream usually took several minutes and were succeeded by 10 or 15 min of conditioning before testing with additional rapid changes.

Temperature was monitored by a small thermistor (Fenwall Electronics, BC 32 L1) 3 mm downstream from the preparation. The time course of thermistor temperature during rapid changes resembled an e-function with a half-time of about 125 ms. End values were reached in less than 1 s. Because the elongated sensillum (Hess and Vlimant 1983a) has a volume 500 times smaller than the thermistor's (roughly a cone 200 µm long with a base diameter of 14 µm vs. an eliptical body 200 by 400 µm), the time course of T within the sensillum is presumably much shorter. No realistic way was found for determinating it, however, in the absence of precise data on the heat capacity and conductance of structures surrounding the thermally sensitive regions. For this reason no values are offered for the average rate of T change there during definite periods after shifting the jets. Instead the difference in the temperature of the air stream (ΔT) was taken as the stimulus parameter. The necessary T values were obtained by two automatic readouts from the thermistor circuit, the first just before switching the jets and the second 1 s afterwards. For the purpose of forming differences in a T range, the thermistor readings were accurate within 0.03 °C. Both the method of calibrating the readout and the

arrangement for switching between the jets were described recently in some detail (Loftus and Corbière-Tichané 1981).

Humidity. Because of the difficulty in obtaining accurate readings of humidity within the air streams without altering its value or disturbing the airflow, *Pw* was not measured while an air stream was playing on the preparation. Before rapid-change stimulation it was set at psychrometrically precalibrated values (accurate within 3%). These values did not tend to drift measurably over periods of several hours. For a description of the apparatus, see Loftus (1976).

Impulse frequencies for fixed periods of time were determined by the impulse interval count of the nearest 0.1 interval during 100, 200, 300, 500, 800 and 1,100 ms, beginning with the first impulse after stimulus onset.

Peak frequency on the other hand was calculated from the shortest overall period of 5 consecutive intervals: *a, b, c, d,* and *e*. The 5 were measured in overlapping blocks of 3 intervals and the mean of each block was formed. The reciprocal of the average of the 3 means was then the peak frequency: $F_{max} = 1/((a+b+c)/3+(b+c+d)/3+(c+d+e)/3)/3)$. Reductively this procedure yields $F_{max} = 1/((a+2b+3c+2d+e)/9)$. It emphasizes the central intervals of the group of 5, where the shortest is usually found, and tends to avoid some of the error involved in measuring single intervals.

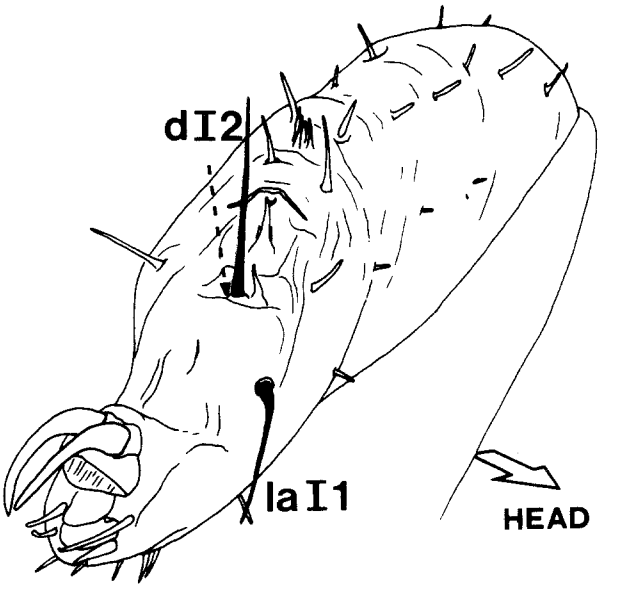

Fig. 1. Tarsus of right foreleg viewed from its distal end. Sensilla recorded from are shown in black. These are innervated by 4 sensory cells, 2 with tubular bodies and 2 extending up into seta. Designations are from previous papers on structure (Hess and Vlimant 1982, 1983). Dotted line indicates position of tungsten electrode

Results

General response to temperature change

An electrode inserted into the base of front-tarsal sensillum dI2 on the side opposite to its small neighbor (Fig. 1) reveals a pair of units distinguishable as a rule by the amplitude and form of their impulses. This pair responds antagonistically to changes in temperature brought about by shifting between constant-temperature air streams. The same two units can be encountered in front-tarsal sensillum laI1. Figure 2 is an example. An air stream at 25.8 °C had been playing on the foreleg of *Amblyomma variegatum* for some minutes. Both units were discharging at about 3 imp/s. Then a second jet at a higher temperature (*T*) but at the same partial pressure of water vapor (*Pw*) was directed at the foreleg for 1.3 s while an attached gate deflected the first stream. *T* rose to 27.0 °C. Impulse frequency (*F*) of one unit quickly rose, peaked at 76 imp/s and gradually diminished. The other unit fell silent. The second jet was then snapped away and *T* rapidly returned to its original level. The unit whose *F* had risen during warming fell silent, while the firing rate of the other mounted and peaked at 26 imp/s, even after such short exposure to a slightly higher temperature.

In addition to shifting between air streams of different temperatures, qualitatively the same antagonistic responses of the two units to rapid cooling or warming can also be elicited 1) by deflecting the air stream with a piece of stiff paper and letting

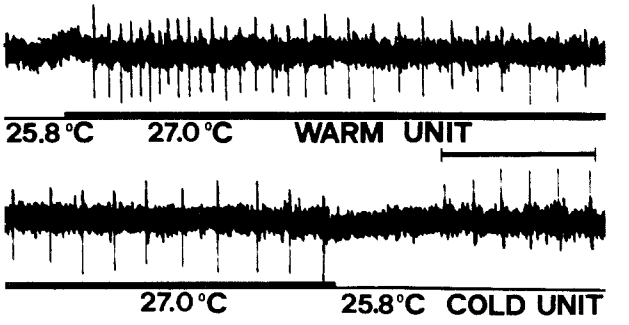

Fig. 2. Extracellular impulses from warm and cold unit in single sensillum. Impulses from the 2 units have opposite polarity here. Time scale: 200 ms. Exposure to warm-air stream: 1.3 s. Conditioning time in cooler air: several minutes

the warmer (or cooler) *still* air of the laboratory contact the tarsus, and 2) by focusing an unfiltered incandescent light source on the tarsus (warming) or by interrupting it (cooling). In each situation the firing rate of one of the units rose while that of the other fell.

Impulses from both the cold and the warm units were not present in all recordings, either from the dorsally located bristle dI2, from which 90% were made, nor from bristle laI1 on the latero-anterior face, the only other sensillum checked for the presence of thermoreceptors. Of the 54 units which held long enough for extensive testing, 33 were cold receptors and 21 warm. In 12 of these cases both antagonists, one cold and one warm,

were encountered in a single sensillum. Some were used for establishing differential sensitivity and resolving power, others to establish modality.

Though no single convincing hypothesis can be advanced to explain the absence of either receptor in recordings from individual sensilla, plausibilities abound. Far fewer sensilla were sectioned than recorded from. Perhaps some dI2 or laI1 sensilla lack the full complement of 4 cells. Faulty electrode shape, positioning, or insertion depth may have been conducive to silencing some. Or an individual sensory cell may not yet or no longer have been functional.

Other modalities

As the absence of wall pores would indicate (Hess and Vlimant 1983a), no unit in either sensillum reacted to any mixture of odorous substances tested. These include aliphatic unbranched alcohols, aldehydes and acids (chain length, 2–12 C-atoms), esters, terpenes and aromatics, all in 10^{-1} and/or 10^{-2} molar solution in paraffin oil.

Neither did any unit respond to rapid reductions or increases in relative humidity with a change in firing rate sufficient to be detected by acoustic or visual monitoring. These changes were usually from 10.6 Torr of water vapor to a value close to zero (drying agent, 96% H_2SO_4), or the reverse. At 22 °C these changes amounted to jumps of 54% relative humidity.

The close passage of a small but strong permanent magnet had no effect either.

Mechanical stimulation

As the presence of two cells with tubular bodies would indicate, two units respond to vibration and bending. Neither discharges spontaneously, but both react to a tap on the heavy steel plate to which the mounting stage is attached. Both also respond when the seta is bent with a glass fiber, one with a train of impulses and the other with a short burst.

That neither mechano-sensitive unit is identical with the warm or with the cold unit was demonstrated by recordings in which the impulses of all four units were clearly distinguishable by reason of spike amplitude and/or form (Fig. 3). Warm unit impulses here were diphasic and only about half as large as any of the others. Cold unit impulses were triphasic with two downward peaks and one upward. Impulses of one mechano-unit were also clearly triphasic, but of opposite polarity (two upward peaks and one downward). Those of the other were more complex, appearing to have

Fig. 3A–D. Four different units in a single sensillum manifest themselves under different stimulus conditions: **A** during warming; **B** during cooling; **C** during bending of the seta; **D** upon its return to neutral position. In recordings from this sensillum warm unit impulses (**A**) tended to be the smallest and of short duration; trace sequence, down-up-down. Impulses from the cold unit (**B**) and from mechano-sensitive unit I (**C**) were usually about the same amplitude but of opposite polarity: trace sequence in **B**, down-up-down-up; in **C**, up-down-up-down (ooo). Though the complex impulses in **D** may be interpreted as coming from a single unit (mechano-II), they seem to be more likely composite forms resulting from the partial superposition of impulses from mechano-I and another unit (●●●). The trace sequence (up-down-up-down-up-down) appears to exclude the cold unit from both ends of these complex forms. Since the mechano-sensitive units do not fire spontaneously or during temperature change, they do not interfere with recordings from either temperature-sensitive unit. The antagonism of mechano-I and II is probably only partial. Time scale and amplification same in **A, B, C, D**

5 peaks (3 upward and 2 downward). These may be composite impulses from both mechano-units with some overlap, in which case both would be triphasic and both of opposite polarity to those of the cold unit.

In about 30% of the sensilla additional impulses were encountered with an amplitude greater than any just described. Their form is unusual with at least 3 downward peaks and 2 upward with tops often flattened. Though not always, in many cases their frequency could be more than doubled by switching on the halogen lamp used during electrode insertion. When one of the heat filters was removed, the warm unit fired too, but its impulses arrived at least 12 ms before the high-amplitude ones, usually much sooner. Warm or cold air streams had no obvious effect on their frequency; neither did bending the seta. The high amplitude of these impulses coupled with electrode position and the rather isolated location of np/C sensilla rendered sensory cells of other sensilla an unlikely source, however. Rather, the complex form and late arrival of these high-amplitude impulses led to the suspicion that the operation to eliminate muscle spiking was not always a complete success. Some efference, indirectly affected by intense light, may have gotten past the partially crushed coxa.

The units dealt with in the remainder of this paper were insensitive to mechanical stimulation but patently so to rapid T change.

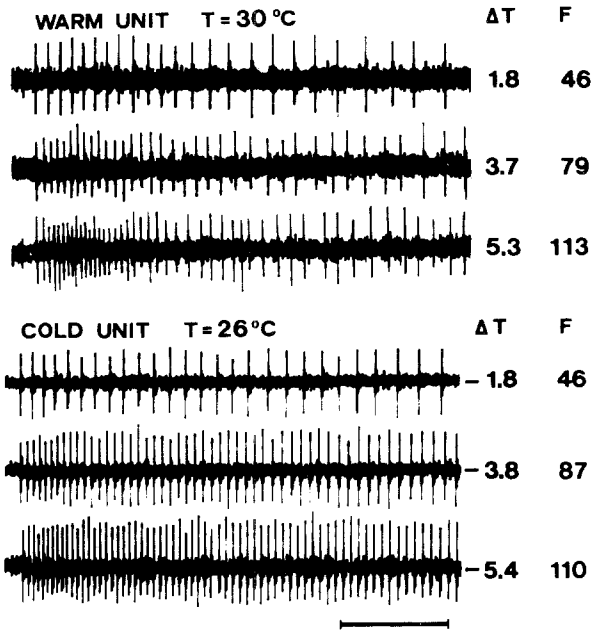

Fig. 4. Impulse trains of a warm and a cold unit in response to abrupt temperature changes ($\Delta T = T_2 - T_1$). Response magnitude for first 100 ms indicated under F. Despite similar time courses of T, individual warm unit responses tend to develop irregularities more quickly than cold. The appearance of single-unit recordings has 2 sources: (1) neither mechanosensitive unit fires without mechanical stimulation, and (2) stimulation by warming or cooling elicits a rapid discharge from one of the thermally sensitive units and silences the other, its antagonist. Time scale: 200 ms. Interstimulus conditioning time at T_1, 3 min. Exposure to T_2, 1.3 s

Table 1. Summary of data used to determine relationship of impulse frequency (F based on first 100 ms of response) to the amplitude of temperature change (ΔT). Relationship here approximated by linear regressions. Their slopes indicate differential sensitivity to ΔT. Values implying variation are means ± standard deviation

Type of unit	Cold	Warm
Units tested extensively	33	21
Units used for characteristic curves	19	7
Number of curbes	27	16
Number of points/curve	12±6	14±5
Initial T	26±4 °C	26±3 °C
ΔT range usually covered	7 °C	7 °C
Slopes of linear regressions approximating relationship of F to ΔT		
Slopes in (imp/s)/°C (b)	−16.1±10.4	17.6±9.5
Correlation coefficient (r)	−0.970±0.038	0.982±0.019

Differential sensitivity to rapid temperature change

In all cells examined quantitatively, rapid T changes ($\Delta T = T_2 - T_1$) were administered by shifting from a reference air stream (T_1) to a second stream (T_2) for 1.3 s and then back to T_1 for a 3-min recovery period. As the spike trains of Fig. 4 would suggest, the reaction of both cold and warm receptors to sudden changes in T is as steadily increasing function of ΔT, with due consideration given its sign. The relationship is exemplified in Fig. 5. Frequency (F) based on the first 100 ms of either cell type continues to rise with the size of the jump, for the first 12 °C quite linearly. To increase the number of characteristic curves per unit, the ΔT range was usually limited to about 7 °C, however.

Basic data are summarized in Table 1. Though the number of cells is not large for either type, the number of tests per characteristic curve is, especially in view of the 3-min recovery periods between tests. Moreover, several cells held up for characteristic curves at different initial temperatures. Of more importance, however, is the quality of the curves. As the correlation coefficients (r) show, the progression of F with respect to ΔT is very orderly. For cold units, r^2 indicates that in a series of tests with rapid T changes, an average of 94% of the variation in F can be explained by a linear regression; for warm units, 96%. When

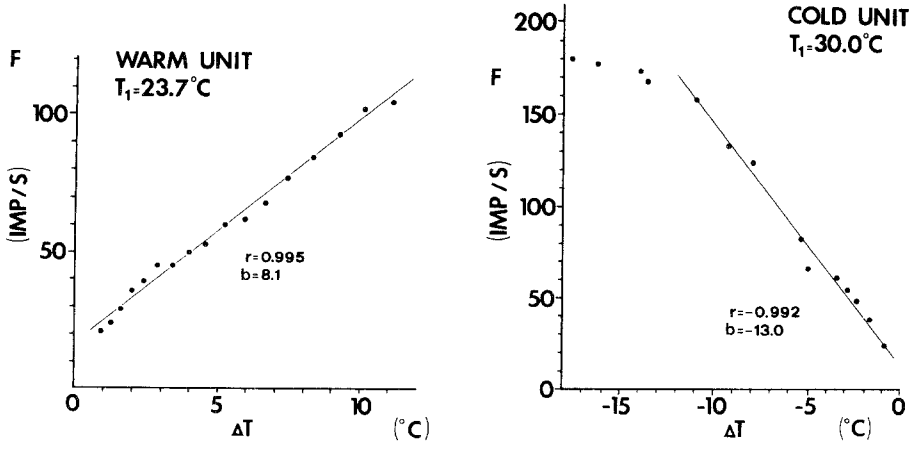

Fig. 5. Characteristic curves of single warm and cold units. F based on first 100 ms of the response. Linear regressions closely approximate the course of the functions throughout the ΔT range tested on the warm unit beyond 0.5 °C, but only for T drops up to 11 °C on the cold unit. The tendency of this function to level off may perhaps be retarded by extending conditioning times at T_1 beyond 3 min. r: correlation coefficient. b: slope of regression. The large difference in absolute slope values of the warm unit from those of the cold was rather unusual

mean r is reduced by its standard deviation, the percentage drops only to 87% for cold unit responses and to 93% for warm.

Despite the high degree of linearity of F as a function of ΔT in both types, the slopes of the regressions display considerable variation. Within the range of their standard deviations, slope values for the cold units can differ by a factor of 4.6; for the warm, 3.3. Thus only limited importance should be attached to the fact that means of both are close. As such they merely provide a rough estimate for slope or differential sensitivity of an as yet unrecorded unit: i.e. that its impulse frequency as determined from the first 100 ms of the response will differ by about 17 imp/s for each degree C that the jumps in temperature differ. The variation in sensitivity may be connected with T_1, the temperature from which the rapid changes were initiated. The number of successful recordings is not sufficient as yet, however, to establish a clear quantiative dependence on it.

Resolving power

Because the relationship of F to ΔT in both types was so orderly, the possibility of determining their resolving power appeared meaningful, despite the small number of characteristic curves. By resolving power here is meant the difference which must separate two stimuli for one of them to be correctly identified with a given probability (e.g. 90%) as being greater than the other. The basis for the identification is a single response to each stimulus from a single cell of average differential sensitivity. The method for determining resolving power described by Loftus and Corbière-Tichané (1981) will be employed here. It requires (1) that the deviations of of individual points from their regressions be normally distributed about a mean of zero, and (2) that the absolute value of the deviations be independent of regression slope.

As regards the second point, the correlation coefficient for the absolute value of the deviations with respect to slope is -0.0794 for cold units and 0.1151 for warm. In neither case is even the sign significant (Sachs 1979). Slope would account for little more than 1% of the extent of deviation, if that.

As regards the first point, the average deviation is below 10^{-7} imp/s in both cases. But the distribution of the deviations is almost certainly not normal (χ^2-test). The density of values near the mean is far too great. The curve is bell-shaped, but its center is too high and it falls off much too rapidly on both sides. With such a distribution, the calculations will tend to underestimate resolving power. But in the absence of a better model, the normal distribution will be assumed for calculation in the awareness that the result should probably be more favorable.

The equation for resolving power is: $\Delta x = \frac{\sqrt{2\sigma}}{|b|} \cdot \Phi_{(\gamma)}^{-1}$, in which Δx is the resolving power in °C, b is the average slope of the regressions, σ^2 is the variance of the individual deviations from their respective regressions, γ is the required probability (90%), and $\Phi_{(\gamma)}^{-1}$ is the inverse of the distribution function of a standardized, normally distributed, random variable. $\Phi_{(0.9)}^{-1} = 1.2816$ (Diem and Lentner 1968, Tables p. 28). σ^2 is estimated by $s^2 = \frac{\Sigma\varepsilon^2}{n-2I}$, in which $\Sigma\varepsilon^2$ is the sum of the squared deviations from the regressions, n is their total number, and I is the number of regressions. n is reduced by $2I$ to obtain the degrees of freedom since for each regression two estimators are required, one for the ordinate intercept and one for the slope. s for the cold unit equals 8.13 imp/s; for the warm, 5.82 imp/s. Substituting these values in the above equation together for those of average slope (Table 1) suggests that the resolving power of the warm unit is superior: 0.64 °C vs 0.90 °C for the cold. These are the differences which, according to this estimate, must separate two stimuli if the larger is to be identified with 90% probability on the basis of the first 100 ms of a single response of a single cell of average sensitivity to each of the two stimuli.

In addition to F as calculated from the first 100 ms of the response, however, Fig. 4 suggests that peak F and also larger segments of the spike trains could serve to convey information regarding stimulus magnitude. Figure 6 is a typical example which shows such to be the case. Whether F is determined from the number of impulse intervals (to be nearest 0.1 interval) during the first 100, 200, 300, 500, 800, or 1,100 ms after the first impulse of the response, the values of F as a function of ΔT display an orderly progression as the absolute value of ΔT increases. As the length of the segment increases however, F tends to become smaller and the slope of the regressions less steep. The scatter of individual points about the regressions, on the other hand, would appear to stay about the same. This observation raises the question of whether resolving power may also remain unchanged no matter whether large or small segments of the spike trains are used to evaluate the response.

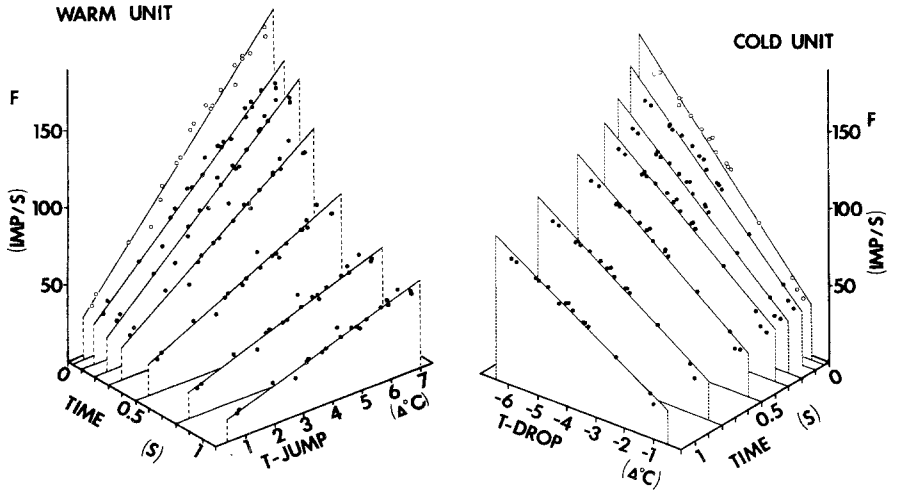

Fig. 6. Effect of the length of the segments of the spike trains used to determine F, on F in response to a given ΔT. F diminishes as the segment is lengthened, and the regression lines approximating the relationship of F to ΔT tend to flatten. This tendency is more pronounced in warm cells than in cold, a situation reflecting spike trains in Fig. 4. For both units $T_1 = 26$ °C

Table 2. Resolving power of the different segments of the spike trains used to indicate response magnitude (F) of cold and warm units. Column 1 (extreme left): type of curve used to approximate the relationship of F to amplitude of temperature change (ΔT). Column 2: length of spike train used to determine F is indicated by subscripts. Segments begin with first spike after stimulus onset, except for F_{max} (peak F) where both the length of the segment and its position in the train vary. Columns 3 and 6: mean slope of characteristic curves. Individual slope values are first derivatives of characteristic curves at values of ΔT actually used as stimuli. Columns 4 and 7: standard deviation (SD) of individual responses from linear regressions or parabolae. Mean slope and SD of responses are variables determining resolving power. Columns 5 and 8: resolving power here is the difference in two abrupt temperature changes, $\Delta(\Delta T)$, discriminable with 90% probability by single cold or warm units at average sensitvity

Characteristic curve approximated by	Response based on	Cold unit			Warm unit		
		Mean slope (b)	SD of responses from curves (s)	Resolving power (°C)	Mean slope (b)	SD of responses from curves (s)	Resolving power (°C)
Linear regression	F_{100}	−16.1	8.17	0.92	17.6	5.82	0.60
Parabola	F_{max}	−18.2	6.42	0.64	19.7	6.97	0.64
Parabola	F_{100}	−16.6	5.33	0.58	16.8	4.86	0.52
Parabola	F_{200}	−15.1	4.15	0.50	16.1	5.05	0.57
Parabola	F_{300}	−14.1	3.86	0.50	12.1	4.59	0.69
Parabola	F_{500}	−13.6	3.12	0.42	8.6	3.48	0.74
Parabola	F_{800}	−12.3	2.98	0.44	6.6	2.46	0.67
Parabola	$F_{1,100}$	−11.2	2.57	0.41	5.9	2.59	0.80

Further, though linear regressions approximate the course of F as a function of ΔT quite well, F for the highest and lowest values of ΔT tends to lie below the lines and F for mid-range values above them. Such is especially true of the cold unit. A higher order curve would yield a better fit. Thus to compare the resolving power of the different-sized segments of the spike trains, parabolae calculated according to the method of least squared deviations were used instead of regression lines to approximate the individual functions.

The use of parabolae required some amendment in determining the values to be entered in the equation above. The other conditions remain the same: that the deviations from the parabolae be independent of slope, normally distributed, and have a mean of zero.

The mean deviation was below 10^{-7} imp/s here too, but the χ^2-test shows the same type of departure from normality as is manifest in the deviations from linear regressions. The concentration of deviations just to either side of the mean is too large: there is a preponderance of small deviations. Thus the method will tend to underestimate resolving power here too.

Since the slope along a parabola varies continuously, no single value could be assigned to the entire segment of the parabola approximating a characteristic curve. Rather, slope values were provided by the first derivative of a given parabola at each ΔT used as stimulus. Thus each deviation had its own corresponding slope. The mean slope for all deviations from all parabolae was entered for b in the equation for resolving power.

No dependence of the absolute deviation on slope could be ascertained. Even the sign of the correlation coefficient was far below the 0.9 level of significance. The above findings apply to both warm and cold cells for F as a function of ΔT, whether F is peak frequency or is determined from the first 100, 200, 300, 5000, 800, or 1,100 ms of the response. To obtain the degrees of freedom in order to calculate the variance of the deviations, the total number of deviations was reduced by 3 times the number of characteristic curves (n-3I), since 3 estimators are needed for the constants which determine a parabola.

The mean slope (b), the standard deviation of the deviations (s), and the resolving power (°C) for F calculated from different-sized segments of the spike trains of both cold and warm cells are given in Table II.

Discussion

Resolving power

Since the interval between impulses in the spike trains tends to grow longer with the passage of time after stimulus onset (Fig. 4), it is evident that the response to a given abrupt temperature change (ΔT) must diminish numerically as progressively larger segments of the trains are used to determine F. Thus it comes as no surprise that the longer this segment is, the flatter the parabola segment tends to become which approximates the course of F as a function of ΔT. But the flatter the parabola, the lower the mean of the slope values along it which correspond to ΔT values actually tested. Under the supposition of spike trains as in Fig. 4, the diminishing values of b in Table 2, columns 3 and 6, are to be anticipated.

What is less evident from simple inspection of the trains in Fig. 4 however, is that the progressive lengthening of impulse intervals after peak F is so regular that the standard deviation of individual responses from the parabolae continues to diminish as the parabolae flatten. The only exception is for $F_{1,100}$ of the warm unit (Table 2, columns 4 and 7). Even less evident is that in the cold unit the rate of diminution of the standard deviation is greater than the rate with which the characteristic curves flatten. Because such is the case, resolving power improves as the segment of the train used to determine F is lengthened between 0.1 and 1.1 s (Table 2, column 5). On the average, resolving power of the cold unit improves by 0.014 °C for each 100 ms the train length is increased, as is shown by a linear regression approximating the relationship ($r=0.84$).

Spike trains of the warm unit, on the other hand, appear less regular. As recorded, the impulse intervals lengthen much more rapidly after peak F and seem less orderly in the way they do it. The standard deviation of the responses from the parabolae diminishes as the segment of the train used to determine F is lengthened, except at $F_{1,100}$ but not fast enough to compensate for the reduction of mean slope. Hence resolving power deteriorates at an average rate of 0.022 °C for each 100 ms the segment of the spike train is lengthened between 0.1 and 1.1 s after stimulus onset ($r=0.82$).

The only other arthropod thermal receptor for which data on resolving power are available is the antennal cold unit of the cave beetle, *Speophyes lucidulus* (Loftus and Corbière-Tichané 1981). If just one such unit of average differential sensitivity is exposed to two successive abrupt drops in temperature from a single starting point, it should be able to identify the larger of the two with 90% probability if one drop is 0.7 °C greater than the other. The estimate was based exclusively on linear regressions as approximations of the characteristic curves, F vs T. On the same basis *Amblyomma*'s tarsal cold receptor could resolve differences in temperature only down to 0.9 °C with the same probability. Yet the correlation coefficient of F with regard to ΔT is much closer to unity in the case of *Amblyomma*. The explanation for the poorer performance of *Amblyomma*'s cold unit is to be sought mainly in its lower differential sensitivity: on the average, −16.1 (imp/s)/°C as against −24.1 (imp/s)/°C for *Speophyes*. In only 2 out of 27 regressions did the gain for *Amblyomma*'s cold unit reach this value. Undoubtedly the estimate for *Speophyes*' cold unit resolving power would have been still more favorable, had the relationship of F to ΔT been approximated by parabolae as was done in the case of *Amblyomma* (Table 2).

Failure to respond to changes in humidity

The fact that neither cold nor warm unit responds to abrupt changes in partial pressure of water vapor eliminates the hypothesis that one or both might be a hygroreceptor. For a hygroreceptor responding for example, to changes in relative humidity brought about by changing the temperature of an air stream should also respond to changes in relative humidity brought about by changing the vapor pressure at a given temperature. (The consideration would of course be complicated if saturation vapor pressure happened to be achieved at some temperature being tested. But this situation was avoided.) The fact, however, that neither

responded to changes in vapor pressure contrasts with the reaction of the cold unit of the cave beetle, *Speophyes lucidulus* (Loftus and Corbière-Tichané 1981). There at least 30% of the cold units responded to an abrupt decrease in vapor pressure, not only to drops in temperature. It was shown that this double sensitivity could not be explained on the supposition that the unit was simply a hygroreceptor. The reaction to cooling was genuinely the reaction of a cold unit, and the additional reaction to abrupt exposure to dry air could be interpreted as resulting from transitory evaporation cooling, also genuine if the cuticle permitted the enthalpic loss of some water, either out of itself if hygroscopic, or from the interior of the larva.

These differences in cold-receptor response to dry air may well reflect differences in cuticle permeability. High permeability may offer little disadvantage in the constant-temperature, high-humidity caves where *Speophyes* is found, but would hardly allow the long periods in open air without nourishment, commonplace for *Amblyomma*. But more may be reflected in differences in permeability than differences in overall thickness, and in those restricted areas which form the covering of sensilla differences may be more specific still. For it must be recalled that the 2 thermal units of *Amblyomma* share a sensillum with 2 mechanosensitive cells, whereas the thermal units of *Speophyes* appear to be housed with a hygroreceptor.

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