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Effects of Temperature on Synaptic Potentials in the Locust Flight System

R. MELDRUM ROBERTSON
Department of Biology, Queen's University, Kingston, Ontario K7L 3N6, Canada

SUMMARY AND CONCLUSIONS

1. Neuronal circuitry in the locust flight system operates normally within a temperature range of 24–42°C. I investigated the effects of temperature on parameters of postsynaptic potentials generated in different neurons following action potentials of the forewing stretch receptor.

2. Increases in temperature reduced latency, time-to-peak and duration (Q_{10}s = 0.51, 0.70, and 0.68, respectively; 24–34°C) and increased the slope (Q_{10} = 1.13; 24–34°) of the excitatory postsynaptic potential (EPSP). However, increases in temperature increased EPSP amplitude below room temperature (Q_{10} = 1.25; 14–24°C) but decreased EPSP amplitude above room temperature (Q_{10} = 0.80; 24–34°C).

3. I conclude that neuronal and synaptic function were affected by temperature in ways predictable by well-established thermal effects on channel conductance and kinetics and on membrane properties. Thus temperature compensation of the output of the flight system must be mediated in some way by the operation of the circuitry.

4. I propose that below room temperature EPSP amplitude was increased by predominant effects on channel conductance and membrane time constant, and above room temperature EPSP amplitude was decreased by a predominant effect on the amplitude and duration of the presynaptic action potential. Further, I suggest that the frequency of the output rhythm is unaffected by the amplitude of single EPSPs, within permissive limits.

INTRODUCTION

Flight motor activity in insects is generated and controlled by circuits of neurons in the thoracic ganglia (Kammer 1985; Robertson 1987). The high power requirements of muscle during insect flight dictate high operating temperatures (Josephson 1981; Stevenson and Josephson 1990), and average thoracic temperatures are often maintained above 40°C (Heinrich 1981). The thoracic temperatures can even exceed the lethal core temperatures (Schmidt-Nielsen 1990) of all mammals, most birds, and many other vertebrates and invertebrates. For example, in honeybees at high ambient temperatures (38°C) the thoracic temperature can reach 48°C without apparently disrupting flight ability (Coelho 1991). Similarly, high thoracic temperatures have been recorded in other bees flying in hot environments (Chappell 1982, 1984). For comparison, the lethal core temperature for man and other eutherian mammals is ~43°C (Schmidt-Nielsen 1990). Flight circuits in the insect thoracic nervous system can therefore function, apparently normally, at relatively high temperatures. Thus insect flight systems provide experimentally accessible neural circuits that operate within a wide range of temperatures, including mammalian body temperatures and higher.

One of the best known neural circuits underlying flight in insects is that of locusts (Robertson 1986, 1989; Robertson and Pearson 1985). During flight there is minimal, if any, regulation of thoracic temperature, and thoracic temperatures are usually in excess of ambient by between 6 and 10°C (Weis-Fogh 1956, 1964). The limits of thoracic temperature for successful flight are 24 and 42°C (Neville and Weis-Fogh 1963; Weis-Fogh 1956), and it is likely that the full extent of this temperature range is experienced by the flight circuitry of locusts under natural conditions at different times. Recently, we have demonstrated that, as in most other flying insects (reviewed in Josephson 1981; May 1981), the wing-beat frequency of tethered flying locusts is increased by raising the ambient temperature (Foster and Robertson 1992). Further, this effect appears to be mediated by an action on the central flight circuitry in the thoracic ganglia, because increases in temperature have an identical effect on the rhythmic output of deafferented preparations (Foster and Robertson 1992). However, the effect of temperature on the frequency of the output of the circuit is mild, with a Q_{10} of around only 1.17, indicating some measure of compensation, automatic or otherwise, for the raised temperature.

Temperature has a wide variety of different, often counteracting, actions on neural circuitry and its operation (Janssen 1992; Montgomery and Macdonald 1990; for reviews). Its effects on synaptic parameters, particularly postsynaptic potential (PSP) amplitude, are notably difficult to predict. Even in locust thoracic ganglia, PSP amplitude has been reported as not changing (Heitler et al. 1977; see also Simmons 1990 for neurons in the locust brain), decreasing (Abrams and Pearson 1982), or increasing (Burrows 1989) in response to temperature elevation. It has been proposed that some of the discrepancy arises from differences in the colonies of locusts used for the experiments (Burrows 1989).

There are at least three reasons for investigating the effects of temperature on the operation of the locust flight circuitry. First, previous investigations (e.g., Robertson 1990, 1991; Robertson and Pearson 1985; Robertson and Reye 1988) were made with the use of semi-intact preparations at room temperature. Investigations at behaviorally and physiologically relevant thoracic temperatures are necessary. Second, because temperature does affect the rhythmic output of the circuitry, it can be used as an experimental probe of the circuitry. Third, the locust flight system provides a model circuit for investigating the effects of high temperatures on neural function; particularly interesting would be those properties or compensatory mechanisms that allow continued operation of the circuitry at temperatures around, or in excess of 40°C.

In this paper I describe the effect of temperatures in the range of 15–40°C on parameters of PSPs in the flight circuitry of *Locusta migratoria*. Although the behaviorally relevant temperatures for the locust flight circuitry are room temperature and above, I also studied temperatures below room temperature, the better to compare my results with those obtained in previous studies (e.g., Abrams and Pearson 1982; Burrows 1989). I used the connections from a peripherally accessible wing proprioceptor, the forewing hinge stretch receptor (SR) (Gettrup 1962), to various flight neurons as model synapses. The wing SR has the advantage of consisting of a single sensory neuron that can be unambiguously identified in extracellular recordings (Robertson 1992). Thus only the postsynaptic elements needed to be recorded intracellularly.

METHODS

Locusts (*Locusta migratoria*) were obtained from a crowded colony maintained in the Department of Biology at Queen's University. The average temperature in the cages was 27°C and the light:dark cycle was 18:6. Males and females were used, and no difference related to the sex of the animal was noticed.

I used a dissected preparation of the locust described previously (Robertson and Pearson 1982). The wings and legs were removed, and the locust was pinned to a cork board and dissected from the tergum to reveal the thoracic nervous system. Nerves 3 and 4 of the meso- and metathoracic ganglia were severed to deefferent most of the flight muscles and thus to help stabilize the preparation. The ganglia were lifted onto a stainless steel plate and held there by the tension in the connectives and the remaining nerve roots. The preparation was grounded to a chlorided silver wire via an Agar/electrolyte bridge. This isolated the ground wire from changes in temperature and prevented the formation of spurious potentials. For some experiments action potentials of one of the forewing SR were recorded from prothoracic nerve 6 with the use of a monopolar silver wire hook electrode insulated from the saline with a mixture of petroleum jelly (Vaseline) and mineral oil. However, at temperatures close to 40°C, the mixture tended to flow away from the nerve/electrode assembly, allowing the saline to short-circuit the electrode to ground. In later experiments the stretch receptor activity was recorded by the use of a glass-tipped suction electrode on prothoracic nerve 6. Activity of the SR was easily recognized, first by the large amplitude of its recorded action potential relative to other sensory activity, second by its spontaneously active firing rate of ~5–10 spikes/s, and third by its pronounced increase in firing rate in response to elevation of the ipsilateral forewing stump (Gettrup 1962). Intracellular recordings were made from the neuropile processes of neurons in the mesothoracic ganglion by the use of glass microelectrodes filled with 1 M potassium acetate (20–60 MΩ). Occasionally, the electrodes were filled at the tip with 4% Lucifer yellow in 0.5 M lithium chloride, and the shafts were backfilled with 0.5 M lithium chloride. This enabled the impaled neuron to be identified by filling it with Lucifer yellow and either viewing it prefixation with an epifluorescence attachment on the binocular microscope (Wild M5A), or preparing it for viewing with a compound fluorescence microscope. For the latter, the ganglia were excised, fixed in 4% paraformaldehyde for >1 h, dehydrated in an alcohol series, and cleared in methyl salicylate. Flight neurons were identified according to standard criteria (Robertson and Pearson 1983).

The thoracic cavity was perfused with a drip flow of standard saline [in mM: 147 NaCl, 10 KCl, 4 $CaCl_2$, 3 NaOH, and 10 *N*-2-hydroxyethylpiperazine-*N'*-2-ethanesulfonic acid (HEPES) buffer] by gravity feed from a 500-ml reservoir. Saline entered the thorax just behind the head, flowed over the thoracic ganglia from anterior to posterior, and exited via a slot cut in the abdominal cuticle. The temperature of the saline in the thoracic cavity was monitored by the use of a copper/constantan thermocouple (BAT-12, Sensortek, Clifton, NJ) located just posterior and slightly lateral to the metathoracic ganglion. To change the temperature of the thoracic ganglia, the saline was heated by the use of a heating coil of Nichrome wire around the Pasteur pipette that directed the flow into the thoracic cavity. The desired temperature was reached by adjusting either the current flowing through the heating coil with the use of a rheostat or by adjusting the rate of saline flow past the heater, usually both. For some of the experiments, the saline in the reservoir was at room temperature so that only the effect of increases from room temperature could be investigated. In other experiments the temperature of the saline in the reservoir was maintained at ~10°C by an ice bath, and temperatures above and below room temperature could be investigated. I allowed 2 min for the temperature of the thoracic ganglia to equilibrate after changing the temperature of the saline. No further changes in the parameters measured were observed after this time.

Standard criteria for presumed monosynapticity of connections were used: processes overlapping in the neuropile; constant latency between presynaptic spike and postsynaptic potential; a synaptic delay of ≤1 ms (at room temperature) after subtraction of the conduction delay; following at high discharge frequencies (e.g., Robertson 1987). There is some immunocytochemical support for the use of these criteria to establish monosynapticity (Robertson and Wisniowski 1988), and in at least one case there is electron microscopical confirmation (Peters et al. 1985). With the use of such criteria, the forewing SR has been found to make monosynaptic excitatory connections with a small subset of flight motoneurons (Burrows 1975) and interneurons (Reye and Pearson 1987) in the mesothoracic ganglion. Most of the excitatory postsynaptic potentials (EPSPs) analyzed here were recorded from depressor motoneurons and from interneurons 302, 313, 507, and 701 as judged by their characteristic physiology. Synaptic parameters were measured from on-line averages of 20–100 EPSPs for each of various temperatures in the 15–40°C range. Data for each EPSP were included in the analysis only if control values at room temperature (~24°C) were recorded and if a return to these control values was evident on return to room temperature. Each successful recording yielded values at room temperature and at least one other temperature.

For 25 successful penetrations in 17 different animals, I measured *1*) latency from the SR action potential to the start of the EPSP, *2*) EPSP amplitude, *3*) time-to-peak of the EPSP, *4*) EPSP duration at half amplitude, and *5*) the maximum slope of the depolarization associated with the EPSP. These measurements were made by hand from 8 × 12–in hardcopy obtained by plotting the stored average EPSPs on a Hewlett Packard X/Y plotter.

The data were normalized with respect to parameter values at room temperature and plotted against changes in temperature relative to room temperature. Exponential and quadratic functions were fitted to the data by the use of the software available in Sigmaplot (Jandel Scientific, San Rafael, CA). This was done merely as a way of indicating trends in the data and does not imply knowledge, or suspicion, of the mechanisms underlying the changes.

RESULTS

Neurons in locust thoracic ganglia are known to receive a continuous barrage of excitatory and inhibitory synaptic input (e.g., Burrows and Siegler 1978; Robertson 1991) of a variety different sizes and from a variety of different sources. This was evident in the experiments reported here, and it was occasionally difficult to detect the smallest EPSPs in the background synaptic activity. Another conse-

TABLE 1. *SR EPSP parameter values at room temperature for 25 different neurons*

	Low Value	High Value	Mean	Q_{10}[a]
Latency, ms[b]	0.47	3.02	1.43 ± 0.6	0.51
Time-to-peak, ms[c]	1.63	10.12	3.24 ± 1.7	0.70
Duration, ms[d]	4.65	29.07	11.50 ± 5.0	0.68
Slope, mV/ms[e]	0.12	3.44	1.16 ± 0.9	1.13
Amplitude, mV[f]	0.28	7.00	2.21 ± 1.7	0.80

Values in Mean are means ± SD. SR, stretch receptor; EPSP, excitatory postsynaptic potential. [a] Q_{10} is given for a 10°C increase from room temperature (~24°C). [b]Latency from extracellular SR action potential to start of EPSP. [c]Time-to-peak of the EPSP. [d]Duration at half amplitude of the EPSP. [e]Slope of the initial depolarization of the EPSP. [f]Amplitude of the EPSP.

quence of this background activity is that the membrane potential and input resistance of the impaled neuron fluctuated constantly, without clear "resting" values (see also Burrows 1985). Thus individual EPSPs could be different in their sizes and shapes as a result of slightly different membrane conditions at the time of their occurrences. Averaging the waveforms triggered from the SR action potential allowed the EPSPs to be detected readily and also eradicated the variations induced by the variable background activity. There was no detectable difference in the parameters of average EPSPs recorded several minutes apart.

The EPSPs that were recorded varied considerably in the values of their parameters at room temperature (e.g., amplitude of individual EPSPs ranged from 0.28 to 7.0 mV in different neurons; Table 1). The low value for the latency (0.47 ms) initially appears impossible given that synaptic delays at room temperature are normally ~0.8–1.0 ms. However, it is important to remember that the extracellular electrode was on prothoracic nerve 6. The SR axon travels from the periphery and branches into mesothoracic nerve 1 (direct to the neurons under investigation) and prothoracic nerve 6. The recording electrode was not in the direct path from the periphery to the impaled neuron but on a side branch. If the extracellular electrode had been further out on this side branch (i.e., closer to the prothoracic ganglion), then it might have been possible even to record negative latencies. Regardless of the initial value of a parameter, the effects of temperature were proportionately the same for each. Sample traces from a single experiment are shown in Fig. 1. The temporal parameters of SR EPSPs were affected similarly with changes in temperature (Fig. 2), although to different degrees. There was a marked reduction in the duration of different components with increases in temperature. Consistent with this was an increase in the rate of depolarization of the EPSP (Fig. 2*E*). An interesting finding was that for temperature increases below room temperature there was an increase in the amplitude of the EPSP, whereas for temperature increases above room temperature there was a decrease in EPSP amplitude (Fig. 2*F*). The peak amplitude was found within 5°C on either side of room temperature.

I confirmed that the EPSPs generated by SR action potentials were representative of other PSPs in the flight system by simultaneously recording intracellularly from a flight interneuron (*501*) and a flight motoneuron (EMn, elevator motoneuron), which have an inhibitory connection between them (*501* to EMn). The temperature effects on the inhibitory PSP (IPSP) recorded in EMn were the same as those on the SR EPSPs (Fig. 3). The presynaptic action potential recorded in *501* showed a dramatic reduction in amplitude and duration with increases above room temperature (Fig. 3).

In this study I did not investigate in a rigorous fashion the effects of temperature on the cellular properties of the impaled neurons. Nevertheless, it was clear that the input resistance decreased with increased temperature. This has since been confirmed for thoracic flight neurons (H. Xu and R. M. Robertson, unpublished observations). Also, after eliminating (by interposing the Agar/electrolyte bridge) the spurious effect of temperature on recorded membrane potential due to potentials developed at the chlorided ground wire, I failed to record consistent differences in membrane potential as a result of temperature changes. Slight changes in membrane potential were noticed, but they could be either depolarizing or hyperpolarizing (for the same neuron in different experiments) and were seldom >5 mV in the range of temperatures from room temperature to 40°C.

DISCUSSION

Like other poikilotherms, locusts are faced with the problem of a body temperature that fluctuates with variations in environmental temperature and with the heat buildup from working muscles. Thus their nervous systems have to operate effectively within a wide range of temperatures. In many ectotherms, locomotor performance is critically dependent on body temperature (Bennett 1990). However, locust flight performance appears to be minimally affected by thoracic temperature, at least within the permissive tempera-

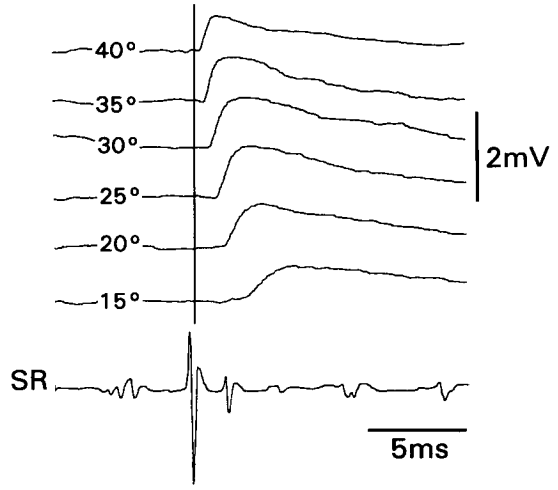

FIG. 1. Effects of temperature on parameters of excitatory postsynaptic potentials (EPSPs) recorded in an unidentified interneuron. The *6 top traces* are each averages of 20 EPSPs triggered from the extracellularly recorded action potential of the stretch receptor (SR; only 1 sweep shown) and recorded at different temperatures (room temperature around 25°C). Temperature is indicated in °C. The vertical line indicates the time of occurrence of the SR action potential in each of the *top traces*. Note that increases in temperature reduce the latency, duration, and time-to-peak of the EPSP; and increase the slope of the EPSP. Note also that the EPSP amplitude is at a maximum around room temperature and that the increase in amplitude below room temperature is coincident with almost all of the increase in slope.

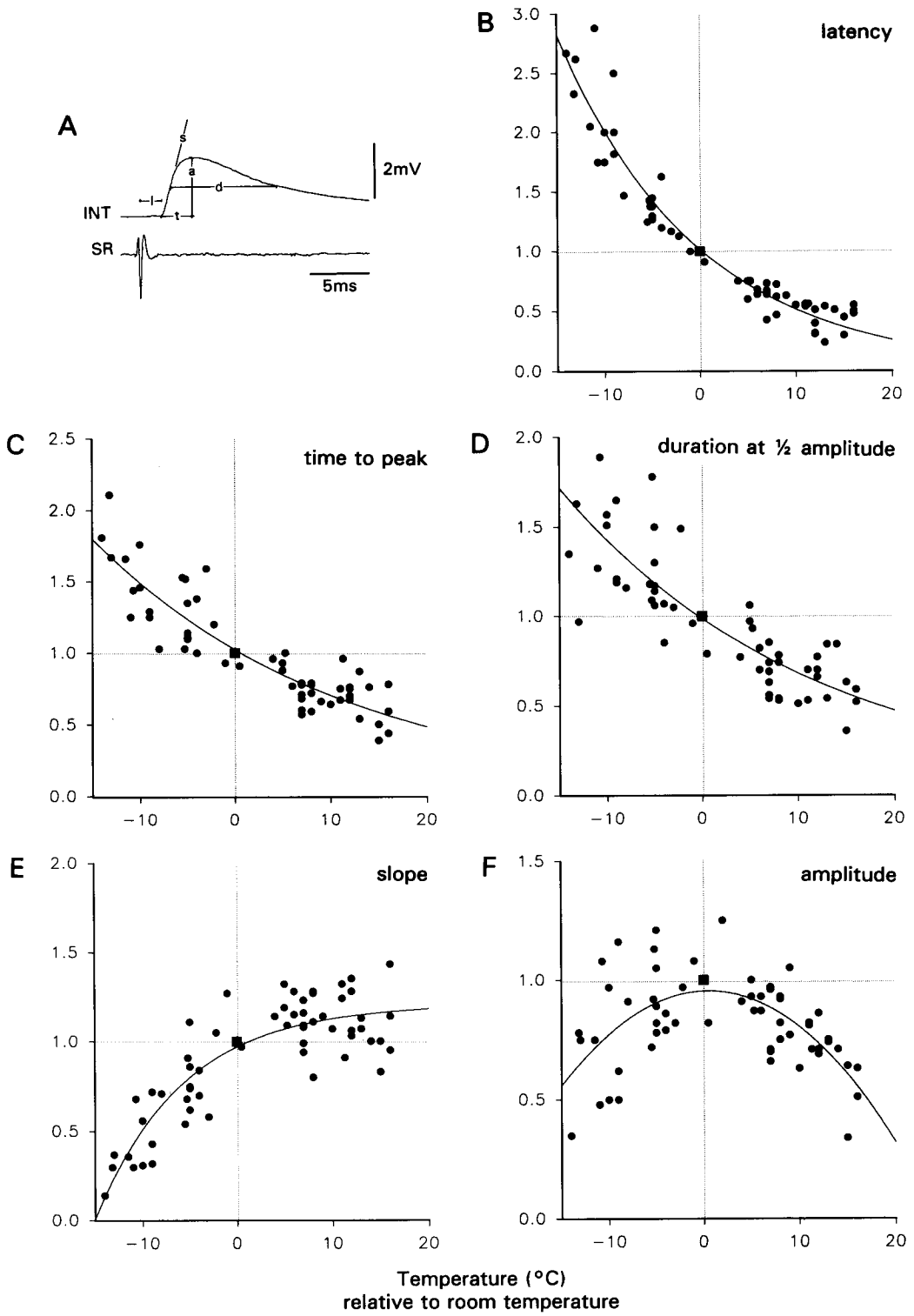

FIG. 2. Effects of temperature on parameters of EPSPs. *A*: parameters of EPSPs measured. Average of 50 EPSPs recorded intracellularly in an unidentified interneuron (INT) in the mesothoracic ganglion following action potentials of a forewing stretch receptor (SR) recorded extracellularly from prothoracic nerve 6, at room temperature (25°C). Sweeps triggered from negative peak of SR spike. a, amplitude; d, duration at half amplitude; l, latency; s, maximum slope of rising phase of EPSP; t, time-to-peak. *B–F*: variation in parameters of EPSPs from SR to mesothoracic neurons as a function of temperature. Temperature was normalized relative to room temperature (~25°C). Each parameter was normalized relative to its value at room temperature. Note that the ordinates on the graphs have different scales. *B*: latency. The line is an exponential fit to the data of the following form: latency = $1.02\, e^{(-0.068\ \text{temperature})}$. *C*: time-to-peak. The line is an exponential fit to the data of the following form: time-to-peak = $1.02\, e^{(-0.038\ \text{temperature})}$. *D*: duration at half amplitude. The line is an exponential fit to the data of the following form: duration = $0.98\, e^{(-0.037\ \text{temperature})}$. *E*: maximum slope. The line is an exponential fit to the data of the following form: slope = $0.24\,[1 - e^{(-0.11\ \text{temperature})}] + 0.97$. *F*: amplitude. The line is a quadratic fit to the data of the following form: amplitude = $0.95 + 0.0015\ \text{temperature} - 0.0017\ \text{temperature}^2$.

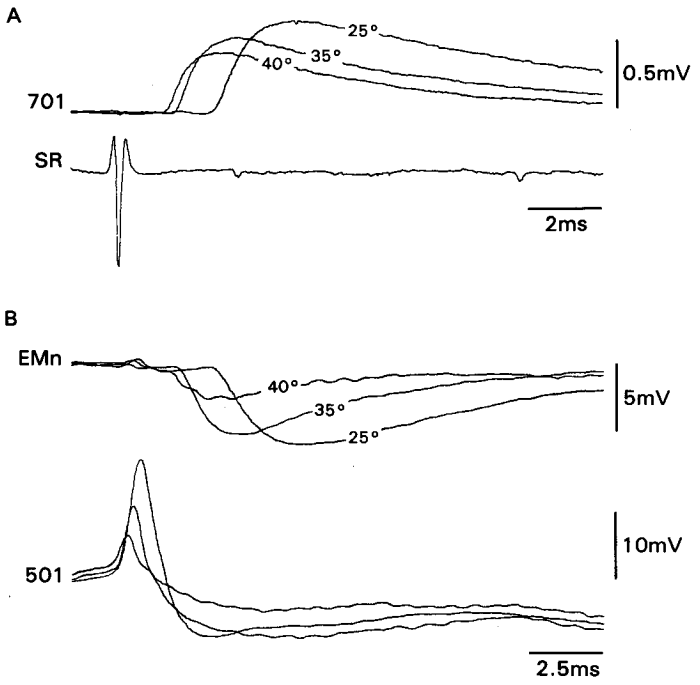

FIG. 3. Similar effects of temperature on the parameters of the EPSP from SR to *interneuron 701* (*A*) and on the IPSP from *interneuron 501* to an elevator motoneuron (EMn; *B*) in the mesothoracic ganglion. Temperature is indicated in °C. *A*: each *701* trace is an average of 100 sweeps triggered from the negative peak of the SR spike. *B*: EMn trace at 25°C is an average of 25 sweeps, the EMn trace at 35°C is of 50 sweeps, and the EMn trace at 40°C is of 18 sweeps; each triggered from the rising phase of the action potential recorded from *501*. Note that the action potential recorded intracellularly from *501* decreased markedly in amplitude and duration with increases in temperature (small spike at 40°C, midsize spike at 35°C, large spike at 25°C; not indicated on figure).

ture limits (Foster and Robertson 1992; Weis-Fogh 1956). The results described here demonstrate that neuronal and synaptic function in the locust thoracic nervous system is affected by temperature in ways similar to other nervous systems, homeothermic or poikilothermic (Janssen 1992). Thus any temperature compensation of the output of the flight system must be mediated by the operation of the circuitry. I have described here some of the effects of temperature on synaptic latency and parameters of PSPs in the flight system of the locust. The temporal parameters of latency, time-to-peak, and duration at half amplitude all reduced with an increase in temperature. Q_{10} in the range 24–34°C was between 0.5 and 0.7 for these parameters (Table 1). In the same range the Q_{10} for the initial slope of the depolarization was 1.13, although the change in slope was greater in the range of temperatures below room temperature. The most interesting finding was that the amplitude of the PSP increased with temperature up to room temperature (Q_{10} = 1.25) but then decreased with a continued increase in temperature to give a Q_{10} in the 10°C above room temperature of 0.80.

PSP amplitude

The rise and fall of PSP amplitude can be attributed to a combination of several different effects of temperature on neuronal properties. Before considering these in detail, it is worth pointing out that the resting frequency of discharge of the SR was only marginally affected by temperature. The effects shown here could not be attributed to facilitation or fatigue of the PSP as a function of the firing rate of the SR. Also, it is unlikely that changes in resting membrane potential can account for the effects on PSP amplitude. Heitler et al. (1977) discovered a 4- to 6-mV/10°C hyperpolarization with heating in locust thoracic neurons [incorrectly reported in Burrows (1989) as an equivalent depolarization with heating), whereas Abrams and Pearson (1982) found no consistent effect on membrane potential. In these experiments I too could not establish a consistent effect. More recently in this laboratory (Xu and Robertson, unpublished observations), we have found a small, variable hyperpolarization with heating of flight motoneurons ($\sim$0.35 $\pm$ 0.25 mV/°C, mean $\pm$ SE, n = 14). However, these changes are too small and variable to have caused any of the alterations in PSP amplitude shown here, although they would undoubtedly have an effect.

The effect of temperature on PSP amplitude described here is particularly interesting, because a consideration of the present result can help to resolve the discrepant results of Abrams and Pearson (1982) and Burrows (1989). Abrams and Pearson (1982) described a decrease of EPSP amplitude in locust thoracic neurons in the range 18–29°C, whereas Burrows (1989) described an increase in EPSP amplitude in a locust thoracic neuron in the range 10–32°C. In Burrows' Fig. 5 it can be seen that most of the increase in amplitude with rising temperature was completed by 22°C, and the relationship is rather scattered, but approximately flat, from 24 to 32°C. In the *inset* to this figure, there is evidence that the amplitude was starting to decrease at 32°C. The results of Abrams and Pearson (1982) were obtained close to room temperature and not in the temperature ranges that have the greatest effect on PSP amplitude. There are two possible explanations for their report of PSP amplitude reduction in the range they investigated. First, the rearing temperature of their experimental animals (23°C compared with 27°C in the present paper) may have influenced the results. If their locusts were acclimated to a lower temperature, it is to be expected that the effects of raised temperature would occur in a lower range (Schmidt-Nielsen 1990). Second, an underestimate of temperature in their experiments, such as has been found in other studies (e.g., French 1982; French and Kuster 1982), would mean that the reduction that they report actually occurred in a higher temperature range. They describe a reduction factor of 1.3 over 10°C. This is equivalent to a Q_{10} of 0.77, which is very similar to the Q_{10} of 0.8 for PSP amplitude reported here.

Consistent with all of the results is the conclusion that small changes of temperature around room temperature have small, unpredictable effects on PSP amplitude in thoracic neurons of locusts, but decreases *and* increases of thoracic temperature of 5°C and more from room temperature cause obvious decreases of PSP amplitude. This conclusion means that during flight, when thoracic temperatures are high, PSP amplitude responds to temperature in the opposite fashion to the way that it responds during much of the locust's other behavior.

At the molecular level increases in temperature will have three primary effects on the postsynaptic membrane that

could affect PSP amplitude. *1*) A moderate increase in single channel conductance (Q_{10} 1.3–1.6) (Correa et al. 1992; Edman and Grampp 1991; Hille 1992; Keynes et al. 1992), consistent with thermal effects on diffusion rates, would increase PSP amplitude. *2*) A larger increase in channel opening and closing kinetics (Q_{10} 3–5) (Edman and Grampp 1991; Hille 1992; Keynes et al. 1992), consistent with thermal effects on protein conformational changes, would decrease PSP amplitude (e.g., Dilger et al. 1991). *3*) Finally, a decrease in membrane resistivity (Q_{10} 0.5–0.75), caused by thermal effects on resting conductances and reflected in decreased input resistances (Burrows 1989; Janssen 1992; Thompson et al.), would decrease length and time constants. In locust thoracic neurons, temperature may also indirectly decrease the membrane resistivity by increasing the rate of synaptic input to the membrane (Burrows 1989). The decreased length constant would tend to reduce the amplitude of PSPs recorded distant from the synaptic site (as is the case here) by increasing spatial decrement. Nevertheless, the decreased time constant relative to the same transient synaptic current would decrease the attenuation of the PSP with distance (Jack et al. 1983) and could more than offset the effect of a decreased length constant to give a larger amplitude PSP.

Presynaptic temperature effects that would affect PSP amplitude include the amplitude and duration of the presynaptic action potential (Burrows 1989; Janssen 1992; Thompson et al. 1985; Weight and Erulkar 1976), which are both decreased with increasing temperature. This was observed in the present experiments (see Fig. 3) but was not investigated in detail. An opposing factor is presynaptic calcium concentration, which could be increased by temperature increasing the presynaptic calcium channel conductance and kinetics, thus increasing the amount of transmitter released (e.g., Charlton and Atwood 1979; Llinas et al. 1987).

It is clear that there are many temperature-dependent processes that could have conflicting effects on PSP amplitude. The results show that the balance of these effects was dependent on the range in which temperature was changed. At the lower temperatures used here, the processes resulting in enhanced PSP amplitude predominated, whereas at the higher temperatures the opposite occurred. I propose, as reasonable testable hypotheses, that *1*) PSP amplitude increased with temperature increases up to room temperature because of predominant effects on increased channel conductance and decreased membrane time constant and *2*) the reduction of PSP amplitude with increases above room temperature was primarily due to the reduction in the presynaptic action-potential amplitude and duration.

Temporal parameters

The slope of the PSP increased markedly up to room temperature with a minor increase after that point. It is likely that this was due primarily to a decrease in the membrane time constant caused by the reduced membrane resistivity (e.g., Lev-Tov et al. 1983), although an increased channel opening rate constant would also have increased the slope. The time-to-peak of the PSP would similarly be affected by the passive properties of the membrane and the kinetics of the ion channel. The decreased duration of the PSP can be attributed to the decreased membrane time constant. The decreased length constant would tend to increase time-to-peak and duration, and decrease the slope by increasing the electrotonic length between the synaptic and recording sites (Jack et al. 1983), so the effect of increased temperature on length constant (via input resistance decreases) seems to have a minimal role.

The latency of the PSP as measured here includes a conduction delay from the extracellular recording site to the active zones of the synapse and a synaptic delay that reflects the release process after the arrival of the action potential. It is well established that both conduction velocity and the rate of transmitter release are increased by increases in temperature (Delaney and Zucker 1990; Janssen 1992). The Q_{10} (25–35°C) for latency described here is 0.51, which would result in a Q_{10} for conduction velocity of 1.96. This is higher than values obtained from a variety of different preparations and a variety of different temperature ranges (Q_{10} around 1.8) (Bullock and Horridge 1965; Janssen 1992). It is also higher than that reported for the cockroach tactile spine (1.75) (French 1985), which agrees well with the value predicted from the Hodgkin-Huxley equations (1.7 in the 10–20°C range and 1.4 in the 20–30°C range) (Chapman 1967; Huxley 1959). This difference may be due to a more pronounced effect of temperature on synaptic delay. In a similar preparation Burrows (1989) has shown that synaptic delay is dramatically affected by temperature with Q_{10}s of 0.17 in the 15–25°C range and 0.5 in the 25–35°C range (my calculation from figures given in Burrows 1989). Thus it appears that the decrease in latency shown here is due to a combination of the effects of temperature on conduction velocity and synaptic delay.

I did not observe conduction failure, even at temperatures of 40°C. For the cockroach tactile spine, production or conduction of the action potential fails at 35–37°C (French 1985). Failure of conduction in the squid giant axon at 33°C (Hodgkin and Katz 1949), or 35–37°C (Chapman 1967), has been attributed to the temperature-induced depolarization for, ignoring such a depolarization, the Hodgkin-Huxley equations predict conduction failure at 44.5°C (Chapman 1967). Interestingly, temperature had minimal effects on membrane potential in the experiments reported here, and any changes observed tended to be hyperpolarizations.

Comparison with effect of temperature on rhythm frequency

The frequency of rhythm generation by the deafferented circuit of flight neurons in the thoracic ganglia has a Q_{10} of 1.19 (25–35°C) (Foster and Robertson 1992). This is a mild effect of temperature on the operation of the circuit, and it represents a Q_{10} on the period of the rhythm of 0.84. As mentioned above, the present experiments have shown a much lower Q_{10} for latency (including conduction and synaptic delays) of 0.51. Put another way, if the rhythm frequency were primarily dependent on delays around the circuit, one would expect it to have a Q_{10} of 1.96 [not the 1.19 observed by Foster and Robertson (1992)]. Thus there must be mechanisms by which the circuit compensates, either automatically or in a controlled fashion, for the effects

of temperature on the temporal parameters of neural function. Mechanosensory neurons innervating cuticular hairs of a similar species, *Schistocerca americana,* are differentially sensitive to temperature depending on their location on the body (Miles 1985), implying the existence of cellular compensatory mechanisms in these sensory neurons. The most temperature sensitive of the sensory hairs are the wind hairs on the head (Miles 1985). However, a first-order wind interneuron, the tritocerebral commissure giant, shows a reduced temperature sensitivity, possibly by virtue of simultaneous, temperature-induced increases in the activity of both inhibitory and excitatory presynaptic sensory neurons (Miles 1992). In an analogous fashion, the decrease in the temporal parameters of single PSPs reported here may be compensated at the point when these single PSPs are integrated to form the compound potential that drives each burst in the rhythm.

Through the range of temperatures in which the frequency of the deafferented flight rhythm increased monotonically, the amplitude of PSPs in the circuit were either at a plateau or decreasing in amplitude. This leads to the interesting conclusion that cycling frequency of the circuit was relatively unaffected by the amplitude of single PSPs, although PSP amplitude may set the limits of frequency (i.e., cycling fails both when PSPs are too slow and weak at low temperatures and when PSPs fail at high temperatures). One possibility is that PSP amplitude indeed has no effect on cycle frequency, and a synaptic connection could serve simply to trigger endogenous mechanisms of burst generation. An alternative possibility is that the decrease in PSP amplitude compensates for the effect of temperature on the conduction and synaptic delays in the circuit. Resolution of this must wait for investigations of temperature on the cellular properties of flight motoneurons and interneurons in the thoracic ganglia.

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