J Comp Physiol A (2005) 191: 281–289
DOI 10.1007/s00359-004-0582-8

ORIGINAL PAPER

Ibrahim Gaaboub · Hansjürgen Schuppe
Philip L. Newland

Position-dependent sensitivity and density of taste receptors on the locust leg underlies behavioural effectiveness of chemosensory stimulation

Received: 1 July 2004 / Revised: 27 September 2004Accepted: 20 October 2004 / Published online: 22 January 2005

Abstract Chemical stimulation of contact chemoreceptors located on the legs of locusts evokes withdrawal movements of the leg. The likelihood of withdrawal depends on the site of stimulation, in addition to the identity and concentration of the chemical stimulus. A significantly higher percentage of locusts exhibit leg avoidance movements in response to stimulation of distal parts of the leg with any given chemical stimulus compared to proximal sites. Moreover, the percentage of locusts exhibiting avoidance movements is correlated with the density and sensitivity of chemoreceptors on different sites of an individual leg. The effectiveness of chemical stimulation also differs between the fore and hind legs, with NaCl evoking a higher probability of leg withdrawal movements on the foreleg. Moreover, sucrose was less effective than NaCl at evoking withdrawal movements of the foreleg, particularly at low concentrations. The gradients in behavioural responses can be partially attributed to differences in the responsiveness and density of the contact chemoreceptors. These results may reflect the different specialization of individual legs, with the forelegs particularly involved in food selection.

Keywords Sensory neuron · Contact chemoreception · Gustation · Insect · Grasshopper

Introduction

Insects detect food and chemicals in their environment by means of contact chemoreceptors (basiconic sensilla) that are small peg-like structures with a single pore in their tips through which chemicals come into contact with the sensory neurons that innervate them (Chapman 1982). Basiconic sensilla are present on most parts of the body of insects but occur in their highest densities on the mouthparts, legs and ovipositor valves (Chapman 1982). In locusts, those on the mouthparts and abdomen influence feeding and oviposition behaviour, respectively (Blaney and Chapman 1970; Ma and Schoonhoven 1973; Simpson et al. 1991; Städler et al. 1995), whereas those on the legs are involved in food acceptance or rejection and in mediating avoidance movements of the legs when they come into contact with chemicals in solution (White and Chapman 1990; Newland 1998; Rogers and Newland 2000).

In locusts, the local circuits that produce and control the movements of the legs are known in considerable detail (Burrows 1996), in particular how they respond to chemosensory stimulation of receptors on the legs and the classes of interneuron that are involved in producing the movements (Newland and Burrows 1994; Newland 1999; Newland et al. 2000). Spiking local interneurons in these circuits receive synaptic inputs from arrays of sensilla on the legs that comprise their receptive fields (Newland 1999). Not all sensilla within the receptive field of an interneuron contribute inputs to that interneuron, and of those that do one area provides the greatest input. Motor neurons that move the tibia and tarsus of the leg have chemosensory receptive fields formed by indirect connection from chemosensory neurons (Newland 1998, 1999). The chemosensory receptive fields of motor neurons, however, differ in one key respect from those of interneurons in that chemosensory stimuli to distal parts of the leg always provide the strongest inputs to the motor neurons (Newland 1998). In this paper, we ask what factors could underlie these properties of the leg motor neurons.

A number of studies have highlighted possible features of the nervous system that may influence the receptive fields of motor neurons but these have not been investigated systematically. Firstly, there is some

I. Gaaboub · H. Schuppe · P. L. Newland (✉)
Southampton Neuroscience Group,
School of Biological Sciences, University of Southampton,
Bassett Crescent East, Southampton, SO16 7PX, UK
E-mail: pln@soton.ac.uk
Tel.: +44-23-80594339
Fax: +44-23-80594459

Present address: I. Gaaboub
Faculty of Agriculture Moshtohor,
Zagazig University, Kaliobia, Egypt

evidence to suggest that the chemosensory neurons that innervate basiconic sensilla on different regions of the leg may have different sensitivities. White and Chapman (1990) showed that the basiconic sensilla on the tarsus produce a higher frequency of action potentials, or spikes, than those on the tibia when stimulated with a given concentration of a salt solution, an observation which could account, to some extent, for gradients in the receptive fields of motor neurons. Secondly, the density of basiconic sensilla may vary in different regions on the legs. Kendall (1970) analysed the number of basiconic sensilla on the legs of *Schistocerca gregaria* and showed that there are more on the prothoracic leg compared to the metathoracic leg. Considering the close proximity of the prothoracic leg to the mouthparts this might implicate the basiconic sensilla on the prothoracic leg with a more specific role in feeding (Dethier 1976; Simpson 1992). White and Chapman (1990) also showed a greater density of sensilla on the tarsus of the foreleg than on the hindleg of *Schistocera americana*. A greater density of sensilla could account for a greater input onto local circuit neurons and thus a systematic change in sensilla density on different regions of a leg could underlie systematic changes in the receptive field properties of the leg motor neurons.

The available data on receptor number and sensitivity of chemosensory neurons are, however, far from complete. This study presents a detailed analysis comparing avoidance movements of the leg elicited by stimulation of different parts of the leg, with the sensory sensitivity and the density of basiconic sensilla. The results are discussed within the context of known input properties of interneurons and motor neurons involved in the withdrawal reflex.

Materials and methods

Experiments were carried out on adult desert locusts, *S. gregaria* (Forskål), approximately 7 days post-moult and of both sexes, taken from our crowded laboratory colony at the University of Southampton. Locusts were reared under a 12:12 h light/dark cycle at 28–30°C and allowed to feed *ad libitum* on a diet of seedling wheat and oats.

Behavioural experiments

To analyse the avoidance movements of the leg evoked by chemicals in a range of concentrations, experiments were carried out in groups of 12 locusts. Five concentrations of a test chemical (NaCl—10, 25, 50, 75 and 100 mM; sucrose—10, 100, 250, 500 and 1,000 mM) and a control of deionised water were used in each experiment, with the presentation order of the solutions systematically altered. Each experiment was repeated five times with new locusts for each replicate, thus there were 60 tests for any given solution. In a separate experiment, the variability in response to proximal and distal leg stimulation was compared for NaCl at a single concentration of 50 mM and for deionised water ($n = 72$ animals). Control tests using deionised water were performed primarily to measure the effectiveness of mechanical stimulation alone in eliciting a response and as a monitor of consistency of response (Rogers and Newland 2000). The droplets containing the test chemicals had a mean volume of 0.04 ± 0.008 ml (mean ± SEM, $n = 40$) so that the mechanical component of the stimulus was always similar (Rogers and Newland 2000). There was an interval of at least 20 min between each presentation of a solution and during that time animals were placed in individual opaque 1 l containers with mesh tops containing a roost site and seedling wheat. During each test, the locust was removed from its container and an opaque hood made from heat-shrink tubing placed over its head to cover the eyes and chemosensory receptors on the mouthparts and antennae (Newland 1998). The locust was then placed on a test arena consisting of a rigid 5 mm mesh nylon sheet, of 200 mm diameter, raised 25 mm above the work surface. Solutions were applied as droplets using a Pasteur pipette held 10–15 mm above a leg (Rogers and Newland 2000). Droplets were only applied when a locust was in a stationary position and the tibia at an angle within ± 30° from vertical. Droplets were applied to sites on the femur and tarsus and ran vertically from the leg and through the mesh, without making contact with the tibia.

All tests were recorded using a video camera (Panasonic WVBP500) mounted on a tripod with a 50 mm lens at 50 frames s^{-1} and recorded on a Panasonic NV-HD680 video recorder for 10 s following the application of the droplets. A time marker (FOR-A video timer) was mixed with the video signal, allowing frame-by-frame analyses of leg withdrawal movements (Rogers and Newland 2000). After each test the leg was rinsed with deionised water and the locust returned to its container. All experiments were performed at 23–25°C and the frequencies of response analysed using chi-square tests and analysis of covariance (ANCOVA) using the statistical software package SPSS.

Physiological recordings

Animals were restrained ventral surface uppermost in modelling clay. The leg of interest was rotated to allow access to particular arrays of basiconic sensilla on the femur, tibia and tarsus. To relate behavioural responses to the neural responses of the chemosensory neurons we recorded the evoked activity of all chemosensory neurons within a sensillum using the tip-recording technique (Hodgson et al. 1955). The total activity of all sensory neurons was recorded since chemicals applied to the leg in behavioural test had the potential to lead to activity in combinations of all of the sensory neurons. Using this technique a blunt recording microelectrode containing the test solution was placed directly over the tip of a receptor. Signals were fed to a standard high impedance DC amplifier and then AC coupled, before being stored on computer following digitisation using a Cambridge Electronic Design (CED) 1401 A/D converter. Neural activity was displayed and printed out using CED Spike 2 software. Spike frequencies were analysed using one-way analysis of variance (ANOVA) and ANCOVA using SPSS software.

A common feature of the responses of contact chemoreceptors is that they adapt rapidly to prolonged or repeated contact with chemical stimuli. To control against such adaptation we analysed the responses of basiconic sensilla to solutions of 75 mM NaCl at different interstimulus intervals from 20 s to 10 min (Fig. 1). With an interstimulus interval of 20 s there was a statistically significant rapid adaptation in action potential, or spike, frequency of sensory neurons innervating the basiconic sensilla (linear regression, $r = 0.27$, $F = 13.84$, $p < 0.001$). The decrease in spike frequency was over 70% at the sixth repetition (ANOVA, $F_{1,54} = 28.3$, $p < 0.001$). No significant decrease occurred with an interstimulus interval of 1 min (linear regression, $r = 0.18$, $F = 1.34$, $p = 0.25$). The changes in spike frequencies of the sensilla with interstimulus intervals of 2 and 10 min were very similar and declined little with repetition number ($r = 0.07$, $F = 0.55$, $p = 0.46$ for 2 min and $r < 0.01$, $F < 0.01$, $p = 1.0$ for 10 min). We therefore chose to use a 2 min interstimulus intervals for all physiological experiments. Physiological data are based on recordings from 215 basiconic sensilla from 42 animals.

To analyse the responses of the chemosensory neurons of the basiconic sensilla to chemical stimulation we used the same five concentrations of NaCl and sucrose used for behavioural analyses. Since responses to 10 mM NaCl were not significant (see Fig. 3a) sucrose was dissolved in 10 mM NaCl to act as an electrolyte. Chemical solutions were applied by placing the recording/stimulating electrode over the tip of a sensillum for 1 s and the frequency of action potentials of all sensory neurones in a sensillum determined.

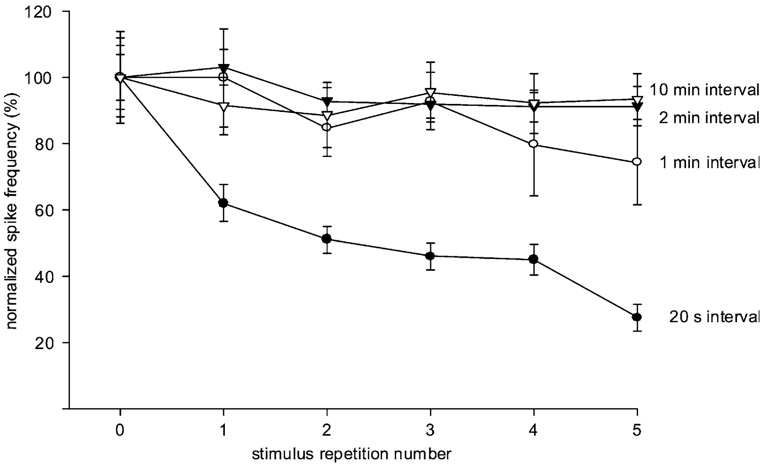
Morphological measurements

The density of basiconic sensilla on dorsal surfaces of the fore and hind legs was determined for femoral and tarsal sites using a Nikon binocular microscope fitted with a *camera lucida* attachment by counting their numbers in rectangular areas of 250 μm^2. Data were based on a measurement from each site from 36 animals for the foreleg and 31 for the hindleg and were analysed using one-way ANOVA.

Results

Chemical stimulation of taste receptors on the legs of locusts evokes an avoidance movement of the leg in which it is rapidly withdrawn from the source of stimulation (Rogers and Newland 2000). During this movement the femur is levated, the tibia flexed and the tarsus levated and frequently held clear of the substrate for many seconds following the stimulus. This behaviour has been quantified in detail by Rogers and Newland (2000). The percentage of animals exhibiting a withdrawal response of the leg has been shown to be dependent on both chemical identity and concentration (Rogers and Newland 2000). We used this simple behavioural paradigm to analyse the dependence of the leg avoidance movement on stimulus location for both

Fig. 1 Adaptation rates of basiconic sensilla on a hind leg. Sensilla located on the dorsal distal femur were stimulated six times with 75 mM NaCl at a given stimulus rate using the tip recording technique. For each sensillum stimulation was repeated at four different stimulus rates and sensory responses measured in terms of overall spike frequency. Here and in Figs. 4, 5 and 6, the stimulus duration was 1 s and spike frequency was determined during the first 0.5 s of the response. *Curves* show the averaged responses (mean ± standard error of the mean, SEM) recorded from 31 basiconic sensilla at stimulus intervals of 20 s (*filled circles*), 1 min (*open circles*), 2 min (*filled triangles*) and 10 min (*open triangles*). Data are normalised with the initial average response equalling 100%



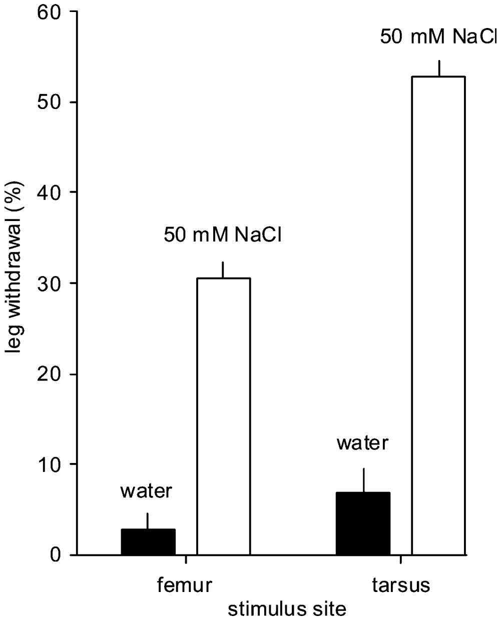


Fig. 2 The percentage of locusts exhibiting a leg withdrawal depends on the stimulus site on the leg. Leg withdrawal movements were evoked by stimulation of basiconic sensilla on different sites on the hindleg with 50 mM NaCl (*white columns*) and water, as a control (*black columns*). The percentage of locusts exhibiting leg withdrawal was higher for sites on the tarsus compared to the femur (mean ± SEM, $n = 72$ locusts)

Fig. 3 Behavioural leg avoidance responses depend on chemical identity and concentration. Animals were stimulated with droplets of NaCl and sucrose solutions at different concentrations. Droplets were applied to the dorsal tarsus. *Curves* show the percentage of locusts exhibiting leg withdrawal movements evoked by stimulation of the fore (*filled circles*) and hind leg (*open circles*) with NaCl (**a**) and sucrose (**b**) (mean ± SEM, $n = 5$ groups). Avoidance responses were strongly correlated with the concentration of a chemical. Note that the higher percentage of withdrawal of the foreleg in response to NaCl stimulation and of the hindleg in response to sucrose stimulation, was conserved across all concentrations

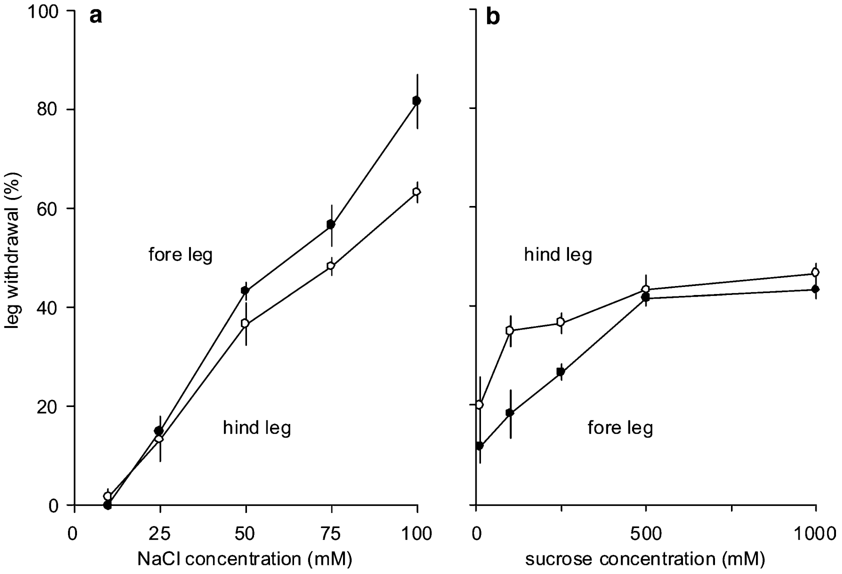
fore and hind legs during stimulation with a range of concentrations of NaCl and sucrose.

Behavioural responses to chemosensory stimulation

The percentage of locusts responding to a chemical solution was dependent on the location of the stimulus site on the hind leg. Applying a droplet of 50 mM NaCl produced a significantly higher percentage of locusts exhibiting a withdrawal response when applied to the tarsus compared to the femur ($\chi^2 = 4.27$, $p < 0.05$) (Fig. 2). Responses evoked by water droplets alone accounted for an overall percentage of less than 7% of withdrawal movements for all animals tested and no significant differences were found between tarsal and femoral sites ($\chi^2 = 1.29$, $p = 0.26$).

The percentage of locusts responding to chemical solutions applied to the tarsi of the hind and forelegs was strongly correlated with the concentration of a chemical, whether it was NaCl (Fig. 3a) or sucrose (Fig. 3b). Increasing the chemical concentration in the droplet applied to the tarsi increased the percentage of locusts producing withdrawal responses to NaCl (ANCOVA, $F_{1,47} = 500.0$, $p < 0.001$) and sucrose (ANCOVA, $F_{1,57} = 83.5$, $p < 0.001$), for stimuli applied to either the fore or the hind legs. NaCl and sucrose were however effective at different concentrations, with NaCl being an order of magnitude more effective at evoking a withdrawal movement at a given concentration. For example, 500 mM sucrose evoked a withdrawal of the foreleg in approximately 40% of tests, while concentrations as low as 50 mM NaCl evoked a similar percentage response of the same leg (Fig. 3a, b).

A comparison between legs revealed that a given NaCl concentration always evoked a significantly higher percentage of withdrawal responses when applied to the foreleg compared to the hind (ANCOVA, $F_{1,47} = 8.43$, $p = 0.006$). When sucrose was applied to the tarsi of both



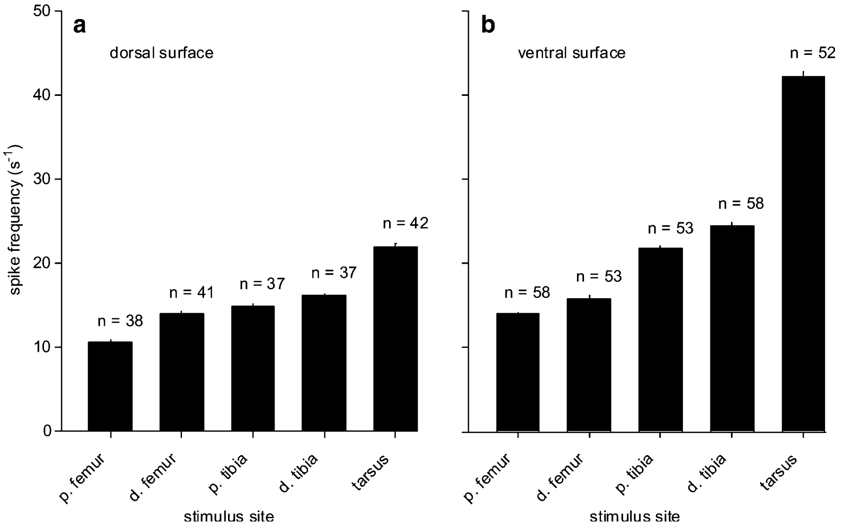


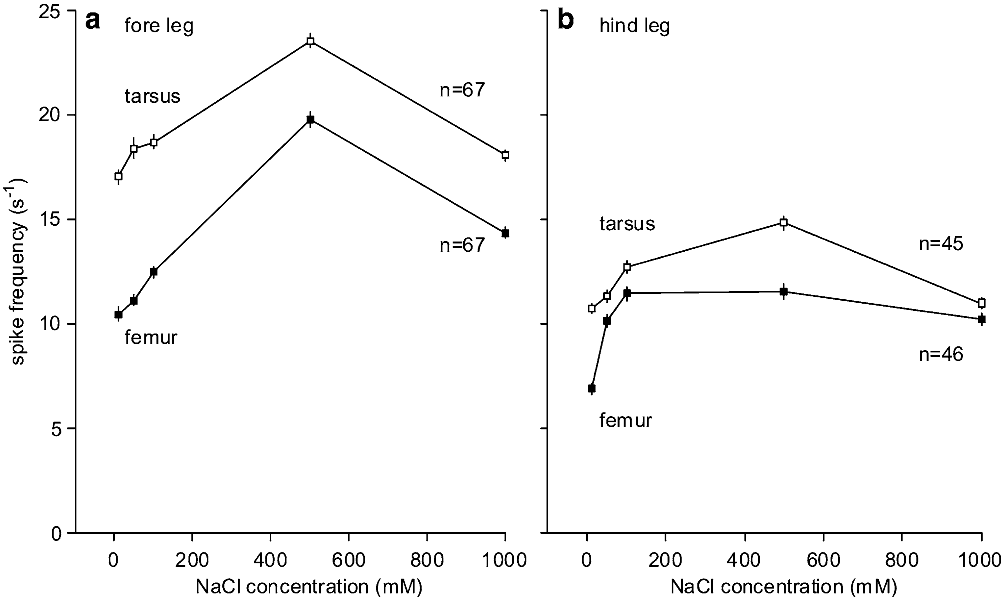
Fig. 4 The sensitivity of basiconic sensilla varies along the length of the leg. Responsiveness of basiconic sensilla located at different positions on the foreleg of each animal was investigated using the tip recording technique. Basiconic sensilla were stimulated with 500 mM NaCl and responses recorded as overall action potential, or spike, frequency. The graph in **a** shows dorsal stimulus sites and that in **b** shows ventral stimulus sites and represents the responses of basiconic sensilla located at five stimulus sites (mean ± SEM; *p* proximal, *d* distal). The sensilla at more distal locations show greater frequencies of spike response

Fig. 5 Effect of stimulus concentration on sensory responses. Basiconic sensilla located at different positions on the fore and hind legs of the same animal were stimulated with NaCl solutions of different concentration. *Curves* show sensory responses of sensilla located on the femur (*filled squares*) and tarsus (*open squares*) of the foreleg (**a**) and hindleg (**b**) for different stimulus concentrations (mean ± SEM). Stimulus concentration and sensory response (overall spike rate) were significantly correlated. At all concentrations responses from sensilla on the tarsus were of higher frequency than those from sensilla on the femur, and sensillar responses on the foreleg of higher frequency than those on the hindleg

legs it also evoked a concentration-dependent increase in the percentage of locusts exhibiting withdrawal responses. In contrast to NaCl, however, sucrose applied to the hind leg evoked a higher percentage of withdrawal responses compared to the foreleg (ANCOVA, $F_{1,57}=9.01$, $p=0.004$). This difference in the percentage of animals exhibiting a withdrawal movement to sucrose was more pronounced at lower concentrations.

Responses of basiconic sensilla to chemosensory stimulation

To analyse the gustatory responses of the chemosensory neurons that innervate the basiconic sensilla on the legs we used the tip recording method (Hodgson et al. 1955) with either NaCl or sucrose (with 10 mM NaCl as an electrolyte) in the recording/stimulating electrode. Since behavioural responses were dependent on the stimulus site on a leg we asked whether changes in sensitivity of



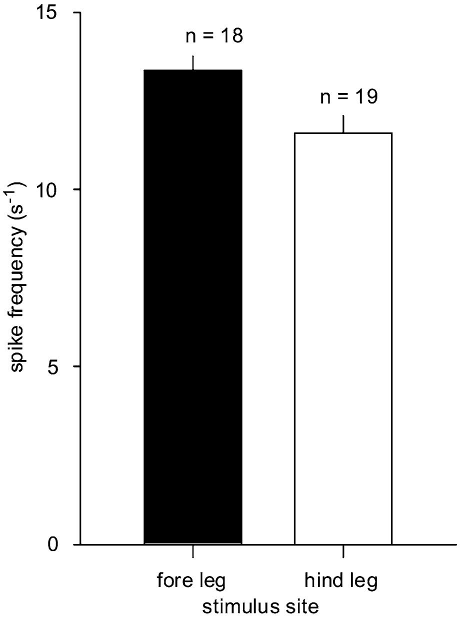


Fig. 6 Differences in sensitivity of basiconic sensilla on the fore and hind legs. Basiconic sensilla on the dorsal tarsus of foreleg (*black column*) and the hind leg (*white column*) were stimulated with 100 mM sucrose and responses measured in terms of overall spike rate (mean ± SEM). Basiconic sensilla on the foreleg showed a higher responsiveness

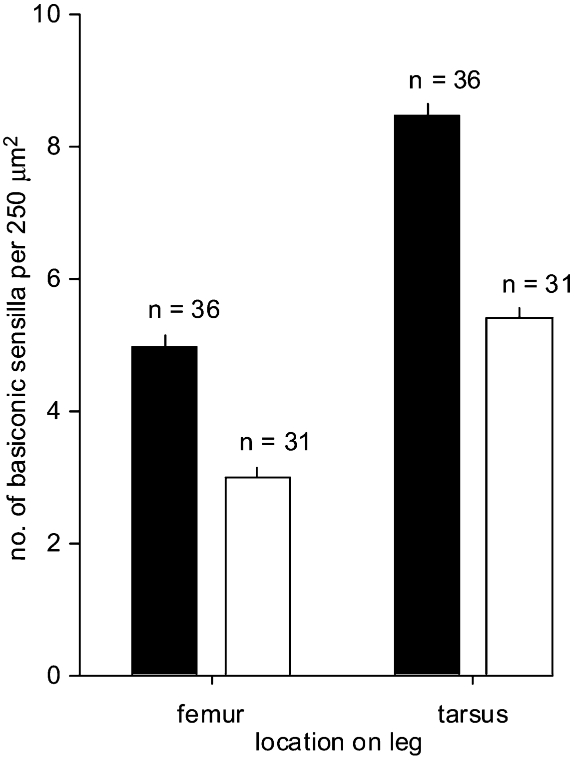


Fig. 7 Density of basiconic sensilla on the leg. The number of basiconic sensilla in an area of 250 μm^2 was determined for several sites on the legs of each animal. Columns show the density of sensilla on the femur (*left*) and tarsus (*right*), both on the fore (*black columns*) and hind legs (*white columns*) (mean ± SEM). Receptor density was higher on the tarsus than on the femur, and higher on the foreleg than on the hindleg

basiconic sensilla also vary systematically along the length of a leg. We chose five sites on the forelegs that included the proximal femur, distal femur, proximal tibia, distal tibia and tarsus, on both dorsal and ventral surfaces and tested the total spike responses of the basiconic sensilla at each site to 500 mM NaCl. The response frequencies of the basiconic sensilla increased with the distal location of the stimulus (Fig. 4), both on the dorsal surface (ANOVA, $F_{4,269} = 179$, $p < 0.001$) and ventral surface ($F_{4,190} = 766$, $p < 0.001$). In each case, the frequency of response of the distal tarsal basiconic sensilla was approximately twice the frequency of the more proximal stimulus sites.

When a range of different NaCl concentrations was applied, basiconic sensilla on the femur were always less sensitive to each chemical concentration compared to basiconic sensilla on the tarsi (ANCOVA, $F_{1,667} = 363.5$, $p < 0.001$ for foreleg and $F_{1,452} = 76.9$, $p < 0.001$ for hind leg) (Fig. 5a, b). For any given concentration, basiconic sensilla on the foreleg (Fig. 5a) showed stronger responses than sensilla on the hindleg (Fig. 5b), irrespective of whether they were located on the femur (ANCOVA, $F_{1,562} = 155.6$, $p < 0.001$) or tarsus (ANCOVA, $F_{1,557} = 640.2$, $p < 0.001$). Similarly, responses of tarsal basiconic sensilla to 100 mM sucrose were greater on the foreleg than on the hindleg (ANOVA, $F_{1,35} = 7.99$, $p = 0.008$) (Fig.6).

Density of basiconic sensilla on the leg

To determine whether changes in behavioural responsiveness were dependent on increased numbers of sensilla we analysed the density of basiconic sensilla at proximal and distal sites on the fore and hind legs. There was a significantly greater density of basiconic sensilla on the dorsal tarsus compared to the dorsal femur on either the fore (ANOVA, $F_{1,70} = 205.7$, $p < 0.001$) or hind leg (ANOVA, $F_{1,60} = 141.9$, $p < 0.001$) (Fig. 7). In addition, the density of basiconic sensilla was significantly higher on the foreleg compared to the hindleg, both on the femur (ANOVA, $F_{1,65} = 93.6$, $p < 0.001$) and tarsus (ANOVA, $F_{1,65} = 152.8$, $p < 0.001$).

Discussion

Gradients in effectiveness of chemosensory inputs

The results of this study show that basiconic sensilla located on distal parts of the leg exhibit a greater responsiveness to chemical stimulation than sensilla on more proximal parts. This observation extends an earlier finding by White and Chapman (1990) who demonstrated that the mean firing rates of basiconic sensilla located on the tarsus were, in general, higher than those on the tibia. Our study shows that the responsiveness of chemosensory sensilla and receptor density are both associated with behavioural responsiveness ($\chi^2 = 13.744$, *df* = 2, *p* = 0.001), suggesting that they could contribute to the receptive field properties of central interneurons and motor neurons processing chemosensory information from the leg (Newland 1999).

Chemosensory neurons form somatotopic maps within the central nervous system (Newland et al. 2000) and spiking local interneurons that receive direct input from chemosensory neurons have receptive fields that map the surface of the leg (Newland 1999). They, therefore, preserve spatial information provided by the contact chemoreceptors. During chemosensory stimulation the amplitude of postsynaptic potentials in interneurons varies in a graded manner along the axis of the leg. Some interneurons are depolarised to a greater extent by chemical stimuli applied to basiconic sensilla on distal parts of the leg and others more by chemical stimulation of more proximal sensilla (Newland 1999). Although sensory neurons at the first stage of processing of sensory information are organised topographically, this is not the case when it comes to the generation of the motor pattern. Chemical stimulation at any site along a leg results in the same motor pattern consisting of an avoidance movement (Newland 1998). This contrasts with the site-specific patterns of movement resulting from mechanical stimulation of exteroceptors on a leg (Siegler and Burrows 1986). Hence, in the case of chemosensory avoidance movements considerable spatial information appears to be lost and what remains are site specific variations in the drive for avoidance behaviour. It is known that motor neurons involved in avoidance behaviour receive their strongest synaptic inputs in response to chemosensory stimulation of distal parts of the leg (Newland 1998). Information about the strength of a chemical stimulus applied to a particular part of the leg is encoded by both sensory neurons and spiking local interneurons and subsequently relayed onto a common set of non-spiking interneurons and motor neurons involved in the generation of avoidance behaviour (Burrows 1996). Consequently, motor neurons would be expected to receive their strongest drive from the stimulation of distal parts of the leg, thus showing the observed non-uniform receptive fields. Our study supports the idea of a strong distal input, provided that leg withdrawal can be taken as a measure for the strength of activation of the motor circuits involved. This assumption seems justified, since the incidence of leg withdrawal depends on the intensity of chemical stimulation (Rogers and Newland 2000, 2002; Fig. 3). The relationship between chemosensory response and percentage of locusts exhibiting withdrawal, however, is not linear. In the case of the foreleg a doubling of the NaCl concentration from 50 to 100 mM causes an increase in action potential frequency of sensory neurons by less than 5%, whereas the percentage of animals exhibiting withdrawal movements increases by almost 100%. This suggests that other, as yet unidentified, factors besides the strength of sensory input are also involved in determining the likelihood of leg withdrawal. These factors could include the increased number of basiconic sensilla and hence increased sensory innervation from distal sites.

What is the function of a gradient in responsiveness along the leg? Avoidance movements of the legs prevent gustatory receptors being in prolonged contact with a chemical stimulus, thus decreasing the potential effects of adaptation. During walking receptors located on distal parts of the leg are likely to be stimulated first and more often than receptors in more proximal locations on the femur. Distal receptors are therefore particularly prone to adaptation, which could explain why withdrawal movements of the leg are observed most often in response to stimulation of distal parts of the leg.

Effectiveness of chemical stimulation differs between fore and hind legs

The relative effectiveness of a given chemical stimulus in producing an avoidance movement not only depends on its chemical identity (Rogers and Newland 2000), but also on which leg it is applied. The percentage of animals exhibiting avoidance behaviour in response to NaCl is greater when the chemical is applied to the foreleg than to the hind leg. To the contrary, sucrose applied to the hindleg produces a higher percentage of responses than when applied on the foreleg. The differences in gradients in responsiveness to sucrose between the legs cannot be explained simply in terms of sensitivity of chemosensory neurons or receptor density alone. In particular, it is possible that receptor density may not be a direct measure for the number of sensory inputs to individual interneurons, since not all receptors need necessarily provide inputs to an interneuron (Burrows 1992). The number of sensilla that provide input to spiking local interneurons within the confines of their receptive fields can vary for different chemical compounds, depending on the sensory neurons activated. Of the tarsal basiconic sensilla investigated in an earlier study only a subset responded to stimulation with 50 mM sucrose, but all fired in response to 50 mM NaCl (White and Chapman 1990). Differences in the relative number of taste sensilla with specific response patterns could represent one of several factors that could account for the stimulus specific variation in responsiveness between fore and hind legs.

During walking chemical stimuli on the substrate provide feedback of potential food sources and potentially noxious chemicals in the environment. Recent studies by Rogers and Newland (2000) showed that the greater the concentration of any chemical stimulus the greater the probability that animals will remove their legs away from a given stimulus, including phagostimulants. The effect of stimulation, differences in threshold and sensitivity of basiconic sensilla on different legs may be related to the specific role of the legs in different behaviours. The forelegs are most likely to make the first contact with a potential food source, or chemical substances that act as a deterrent, and thus might be specifically tuned to the evaluation of food. This implies that the animals would show a tendency to move away from aversive substances (White and Chapman 1990), but stay in contact with a potential food source, as long as this does not compromise the sensitivity of taste receptors in terms of adaptation. It may, therefore, be expected that the forelegs show particularly low avoidance behaviour for sucrose which generally acts as a phagostimulant (Bernays and Chapman 1978) and strong withdrawal in response to high concentrations of NaCl, which can act as a feeding deterrent (Chapman 1977). Our results provide both physiological and behavioural evidence to suggest a specific role of the foreleg in food detection. High concentrations of both chemicals (100 mM NaCl, 1,000 mM sucrose) caused similar withdrawal frequencies when applied to the hindleg, but responses were clearly differentiated for the foreleg with NaCl twice as effective as sucrose, pointing to its role as a food component.

Comparison with chemosensory processing in vertebrates

Our understanding of the organization of the gustatory pathways of insects may provide a greater understanding of how topographical information is relayed to local neural circuits that organise reflex behaviour. In vertebrates, the termination fields of nerves carrying taste information from the oropharyngeal cavity to the nucleus of the solitary tract (NTS) show a distinct topographic segregation (Di Lorenzo 2000) and stimulation of taste receptors triggers vital reflexes such as swallowing (Kajii et al. 2002; Kitagawa et al. 2002). The locust nervous system allows us to understand how a gustatory reflex is generated at a local level and how it may be modulated depending on the context in which chemosensory stimulation takes place. In the locust, chemosensory information is processed by a population of local interneurons (Newland 1999) that contain the transmitter γ-aminobutyric acid (Watson 1986) and provide inhibitory output to interneurons and motor neurons (Burrows 1996). The context dependency, mediated by inhibitory interactions, is thought to contribute to the dynamic tuning of responses to chemical stimuli (Christensen et al. 1998; Di Lorenzo et al. 2003). In vertebrates, gustatory processing has been described as dynamic and distributed (Katz et al. 2002), making it hard to assess the contribution of individual taste responsive neurons to final behavioural decisions. Studies on small neural networks such as those of insects can focus on identified sets of neurons and promise to shed light on common principles underlying the generation of specific taste-evoked behaviours.

Acknowledgements This work was supported by an award from the BBSRC to PLN. We are grateful to Ed Hunt, Paul Yates and Sarah Young for their comments on an earlier version of the manuscript and to Dr Steve Rogers for numerous discussions and advice on statistical analysis. The experiments comply with the "Principles of animal care", publication No. 86-23 (revised 1985) of the National Institute of Health (USA) and with current UK laws.

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