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Chemosensory and Behavioural Adaptations of Ectoparasitic Arthropods

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With 11 Figures and 1 Table

Abstract

Vertebrates respond to being fed upon by ectoparasitic arthropods through their immune responses, grooming and mobility. Arthropod ectoparasites have developed, among others, a series of chemosensory and behavioural adaptations that are crucial to their resource tracking capabilities on vertebrates. Olfactory receptors in chemosensilla on appendages such as the antennae of insects and the first leg-pair of ticks permit these ectoparasites to find hosts from a distance. These include receptors for common respiratory products such as carbon dioxide and other breath components. Ticks, triatomine bugs and phlebotomine sandflies all possess receptors for volatile products common to vertebrates such as branched short-chain fatty acids and aliphatic aldehydes. Both tick and insect ectoparasites signal their presence on vertebrate hosts through the release of pheromones from exocrine glands. These species specific products serve to increase the apparency of the host to conspecifics and enhance encounter between sexes for mating. Constituents of sweat and skin lipids common to vertebrates affect the host attachment responses of arthropod ectoparasites. Receptor cells for these products reside in contact chemosensilla on the legs and mouthparts and these appendages are brought regularly into contact with host's surface as the ectoparasite searches for an attachment site from which to feed. The evidence suggests that evolution has forged a chemosensory disposition that is common to haematophagous arthropods, in which the primary chemostimulants are key by-products of host metabolism or host-associated microflora.

Zusammenfassung

Die Sinnesorgane ektoparasitischer Spinnentiere und Insekten befähigen diese Wirte, auf Distanz zu orten, geeignete Saugorte auf dem Wirt aufzusuchen, sich zu paaren und in der nicht parasitischen Phase geeignete Refugien zu finden.

Anhand verschiedener ektoparasitischer Arthropoden, die Gegenstand unserer Forschung sind, werden Spezialisierungen bei Geruchs- und Geschmackssinn, Sehsinn und Temperaturwahrnehmung im Detail behandelt.

Insbesondere Zecken, Raubwanzen, Tsetsefliegen und Sandmücken nehmen mit Hilfe von Geruchsorganen die Duftausscheidungen ihrer Wirbeltier-Wirte über Distanz wahr. Diese blutsaugenden Parasiten reagieren auch auf visuelle Signale und Wärmereize.

Auf dem Wirt oder in unmittelbarer Nähe verströmen diese Arthropoden ihre eigenen artspezifischen Duftsignale aus speziellen Drüsen, die allein oder in Kombination mit den Duftausscheidungen ihrer Wirte die Wahrscheinlichkeit der Paarfindung steigern.

Die Wahl der Saugorte wird von Geschmacksstimuli beeinflußt, die die Fixierung und das Blutsaugen fördern.

In der nicht parasitischen Phase werden geeignete Eiablageorte und Refugien von den Parasiten mit chemischen Botenstoffen markiert.

Haematophagous arthropods successfully exploit vertebrate hosts through a combination of physiological, behavioural and morphological adaptations (KIM 1985). The classic example of a physiological adaptation is the synchronised life-cycle of the flea with its rabbit host such that larvae and adults emerge when rabbit siblings are delivered (ROTHSCHILD 1965). Vertebrate evolution and adaptive radiation in terrestrial arthropods associated with them underpinned the development of similar life history traits and behavioural adaptations in ectoparasitic groups within the Arachnida and Insecta. Some mites and fleas reside permanently on the host, whereas some hard and soft tick species and hippoboscid flies can exploit the resource from the confines of the host's nest or abode. Selection factors bearing on the parasitic mode of life among arthropods is such that residency time on or near hosts varies between classes and orders of arthropods, and even between members of the same family. Among amblyommid ticks, some are permanent residents of lizards and their refuges (CHILTON and BULL 1991) whereas others such as adult *Amblyomma variegatum* run after large mammals. Likewise, among pupiparous flies of the Cyclorrhapha, members of the Hippoboscidae live in close association with hosts and are sometimes wingless, whereas some Glossinidae track herds of large mammals and require only brief but regular contact with the host for a blood-meal.

The range of vertebrate species occupying diverse habitats has contributed to species radiation in arthropod ectoparasites. The diversity of riverine and savannah tsetse species testify to the role of the habitat exploitation by vertebrates on the African continent on radiation in *Glossina*. Other contributory host factors include immune responses and the propensity of vertebrates to defend themselves against ectoparasites through grooming. Interactions between ectoparasites on the same host further contribute to ectoparasite evolution through niche partitioning (WAAGE 1979). Exploitation by different tick species of predilection sites such as the ears and escutcheon, not so accessible to the host during grooming, underlies the effects of vertebrate defence on the evolution of specific behavioural preferences of these ectoparasites on hosts. Patchy (overdispersed) distributions of ectoparasites on the one hand and host mobility on the other contribute to selection pressures for appropriate sensory and behavioural adaptations on the part of arthropod ectoparasites to ensure a blood-meal. Such adaptations include host finding from a distance, attachment for a blood-meal, feeding and reproduction. This contribution to the symposium recounts examples from our own research on the sensory and behavioural responses of tick and insect ectoparasites to chemostimuli from both hosts and conspecifics that are crucial to their »resource tracking« (KETHLEY and JOHNSTON 1975) capabilities on vertebrates.

Although most of this contribution will be devoted to chemosensory adaptations, evidence abounds for the use of sensory modalities other than chemical by ectoparasitic arthropods. Early attempts to control tsetse flies in Africa made use of visual targets for trapping out these flies *en masse* based on the ability of tsetse to pick out certain visual features in the environment when in search of a blood meal (MCKELVEY 1975). Even hunter ticks such as *Hyalomma dromedarii* equipped with a rudimentary visual system consisting of a lens fixed over a small field of photoreceptors can home in on certain silhouettes (KALTENRIEDER 1990). Heat radiation from the host can provide an orientation cue to blood feeding arthropods. To profit from this in host finding, ticks possess thermoreceptors on the tarsi of the first leg pair that they wave in the air during active search behaviour (HESS and LOFTUS 1984), and night-active triatomine bugs can discriminate between objects at different temperatures (LAZZARI and NÚÑEZ 1989). Male culicine mosquitoes in search of a mate employ the tone of the female wing-beat (ROTH 1948).

2. The Chemosensory Apparatus

Both insects and ticks carry their chemosensory receptor cells in specialized cuticular structures that show a high degree of conservancy across the Arthropoda. A modified section of cuticle or sensillum encloses the distal extensions or dendrites of bipolar sensory cells (Fig. 1). Olfactory sensilla are found on antennae and palps of insects, and in Haller's organ on the first leg-pair tarsi of ticks (Fig. 1 *A*). These appendages are actively moved to sample the air when the arthropod is stimulated with odours (TANEJA and GUERIN 1995, McMAHON and GUERIN 2000). Olfactory sensilla possess numerous pores or grooves in the walls through which air-borne molecules pass (Fig. 1 *B*). In contact chemosensilla, by contrast, a single opening is confined to the tip of the shaft – termed the terminal pore. Generally, these chemosensilla are strategically placed on the tips of legs and palps of arthropods (Fig. 1 *C*) and on the antennae of insects. Such sensilla are brought regularly into contact with the substrate as the organism walks (KRÖBER and GUERIN 1999), and both palps and tarsi are used by male ticks to actively sample the female cuticle during mating (FALK-VAIRANT et al. 1994, DE BRUYNE and GUERIN 1998).

Within the chemosensillum, stimulants encounter receptors on the dendrite membrane of the sensory cells that extend into the sensillar lumen (Fig. 1 *B*, *D*). The number of sensory cells per sensillum can vary from two to several dozen. The dendrites, branched or unbranched, bathe in the sensillum lymph, isolated from the haemolymph by shield cells. The sensillum lymph is rich in odour binding proteins and cations, the latter secreted by the auxiliary trichogen and tormogen cells (Fig. 1 *B*, *D*). A high resting potential of some 150 mV is maintained across the dendritic membrane at rest. The sensory cell depolarises following stimulation through the opening of ion channels in the dendritic membrane. The resulting receptor potential serves to generate action potentials (Fig. 2) that travel along the axons of the sensory cells to the brain (Fig. 1 *B*, *D*). This occurs primarily *via* the antennal nerve in insects and the pedal nerve in ticks. The nature or quality of a chemical message is deciphered by the perceiving organism through the selective response of each sensory cell to a specific stimulus molecule or family of molecules, whereas dose is coded for by receptor potential amplitude and action potential frequency (Fig. 2).

3. Host Odour Perception

Our studies in this area started with an investigation on the effects of volatiles from host animals on the behaviour of the tropical bont tick, *A. variegatum*, a tick with a hunter host-finding habit in the adult stage. Work by HESS and VLIMANT (1986) had shown that this metastriate tick possesses 19 olfactory sensilla on each of the first leg tarsi. We first aimed at characterising the responses of the hundred or so olfactory receptors housed in these sensilla. For this, peripheral olfactory receptors were employed as biological detectors in gas chromatographic (GC) analysis of host volatiles collected from breath, stables, animal rooms, and steer skin washes (see Fig. 5 for method). Apart from host volatiles such as CO_2 (Steullet and Guerin 1992a) and H_2S from the rumen and oral cavity (Steullet and Guerin 1992b), this tick also has receptors for aromatic aldehydes, lactones and phenols, as well as for short-chain fatty acids and aldehydes such as methylpropanoic acid and nonanal (Steullet and

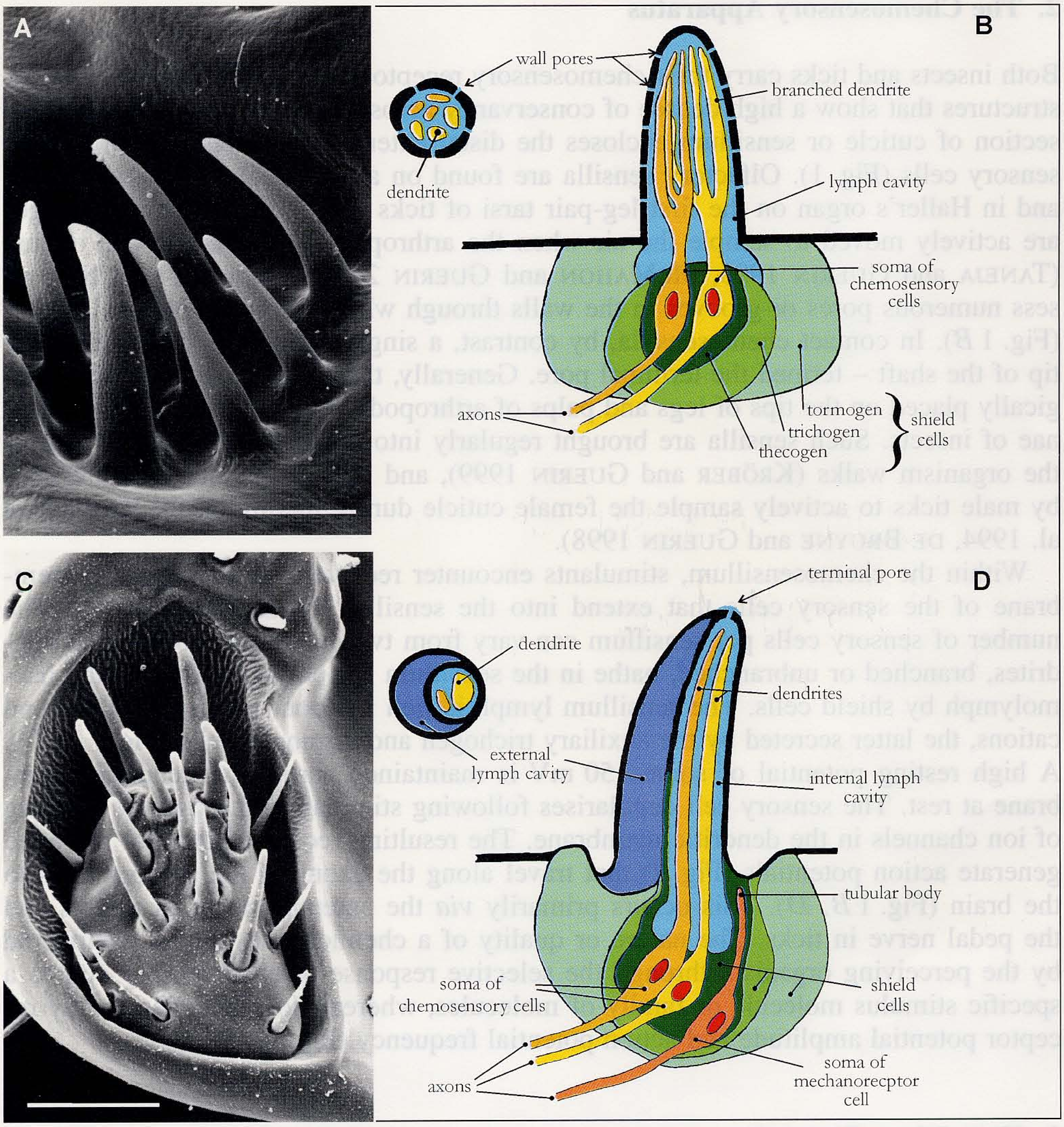

Fig. 1 Olfactory wall-pore sensilla in ticks are concentrated in the Haller's organ as shown for the capsule sensillae of *Ixodes uriae* (A, scale bar 10 µm). Olfactory sensilla of arthropods (*B*) have numerous wall pores through which odour molecules pass into the sensillum lymph (blue). Branched dendrites increase the sensory cell surface provided for receptors such that very low numbers of odour molecules can be converted into action potentials (see Fig. 2). Taste sensilla are concentrated on the appendages of ticks as shown on the palp sensilla of *Ixodes ricinus* (C, scale bar 20 µm). In contact chemosensilla of arthropods (*D*) the stimulating molecules can pass *via* the terminal pore into a lymph cavity containing unbranched dendrites of sensory cells. A second dendrite-free channel is situated either concentric or parallel to the first. Three cells common to both types of chemosensilla (thecogen dark green, trichogen medium green and tormogen light green) envelop the soma of the sensory cells. Mechanoreceptor cells form part of the general configuration in taste sensilla (modified from Maes 1990).

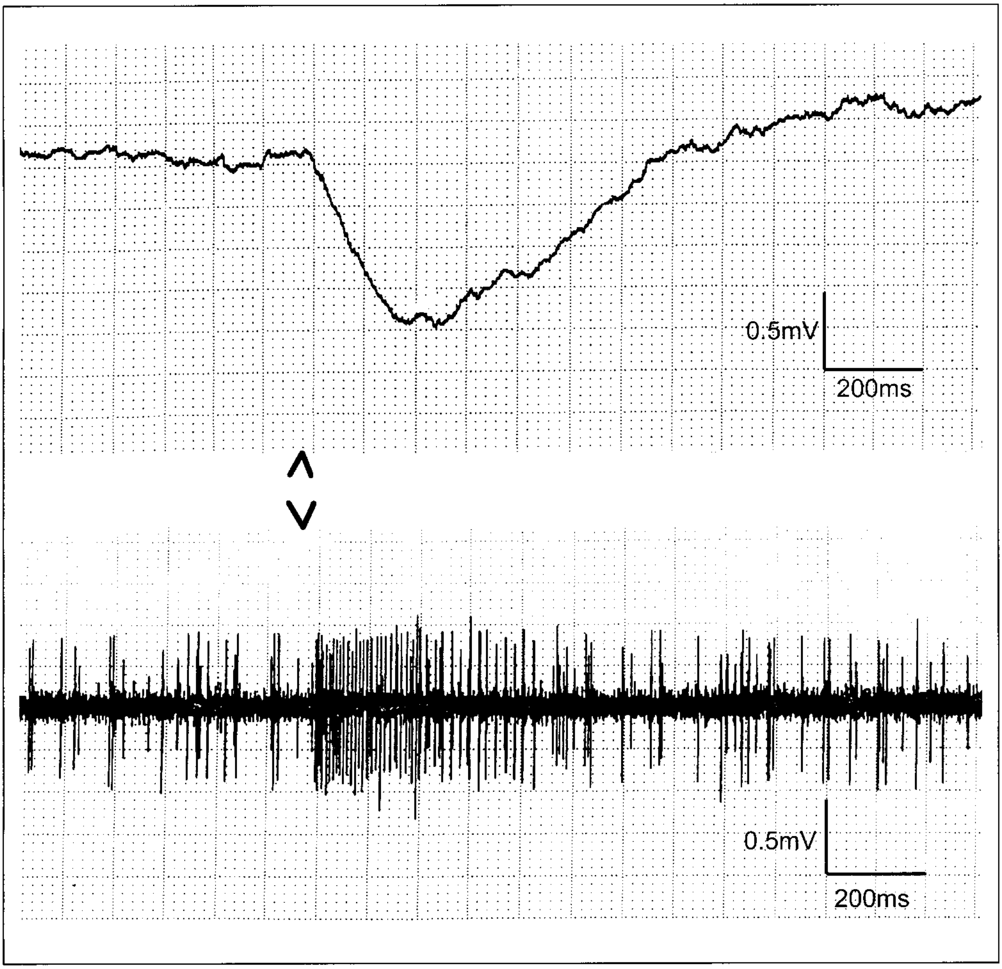

Fig. 2 Receptor potential (upper trace, low pass filtered) and action potentials (lower trace, high pass filtered) recorded simultaneously from a chemoreceptor in a grooved peg olfactory sensillum on the antenna of a 5th instar *Triatoma infestans* nymph in response to stimulation with diethylamine. The stimulus (10 µg-source dose) was held on a filter paper strip in a 5 ml plastic syringe from which 1 ml was displaced in 1 second (at arrow) into an airstream flowing over the electrophysiology preparation (see STEULLET and GUERIN 1992a).

GUERIN 1994a,b). Since then, we have studied the response of *Amblyomma* to the identified chemostimulants in host odour using a servosphere (Fig. 3; KRAMER 1976, MCMAHON and GUERIN 2000). Adult *A. variegatum* of both sexes walk upwind to 10 ng source doses of individual host volatiles (MCMAHON 1999). The most consistent upwind responses are obtained to human breath (Fig. 4) and to mixtures of volatile chemostimulants of different functionality (MCMAHON 1999). Breath contains water and carbon dioxide in addition to many other volatiles such as acetone and H_2S. Both CO_2 (STEULLET and GUERIN 1992a) and acetone (MCMAHON 1999) attract *A. variegatum*. Clearly, despite the limited number of olfactory receptors ticks possess compared to insects, they can discriminate for an array of host volatiles.

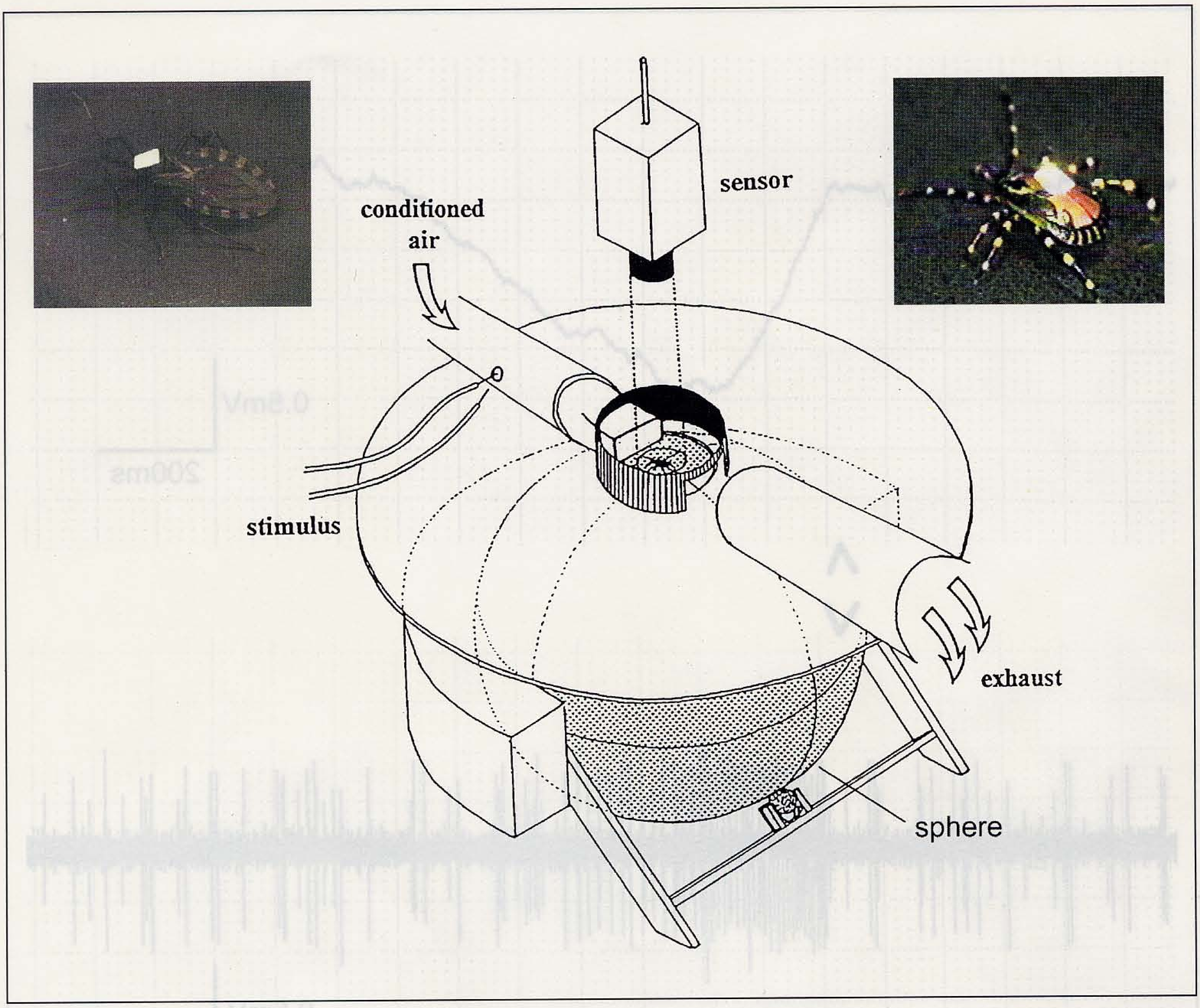

Fig. 3 The set-up for behavioural studies consisting of a locomotion compensator (a perspex servosphere linked to a position sensor), a conditioned air stream for stimulus delivery and an exhaust air flow. Photographs show *Triatoma infestans* (left) and *Amblyomma variegatum* (right) with retroreflectors for detection by the position sensor.

To compliment our work on ticks with an insect ectoparasite we have chosen the triatomine bugs *Triatoma infestans* and *Rhodnius prolixus*, vectors of Chagas' disease. These insects lend themselves to studies on behavioural physiology of ectoparasites as they are readily reared in the laboratory, possess relatively large antennae with a range of chemosensilla types, and the different life-stages are amenable to behavioural tests on the locomotion compensator (Fig. 3). Only adult triatomines possess wings but these can be readily taped together. Triatomines are night active, occupying sylvatic and peridomestic niches. They seek out birds, mammals or man and his domestic animals for a blood-meal from nearby refuges. In the absence of other sensory inputs such as thermal and visual stimuli on the locomotion compensator, triatomines orientate and walk upwind to host odour, its ubiquitous constituent CO_2 and stale rabbit urine (TANEJA and GUERIN 1995). The bugs actively sample the air with their antennae during turning responses towards the odour source. As with the olfactory receptors of ticks, we have characterised the response specificities of triatomine antennal receptors by using them as biological detectors in gas chromatographic analysis of host odour (Fig. 5). As with ticks (above), olfactory receptors of triato-

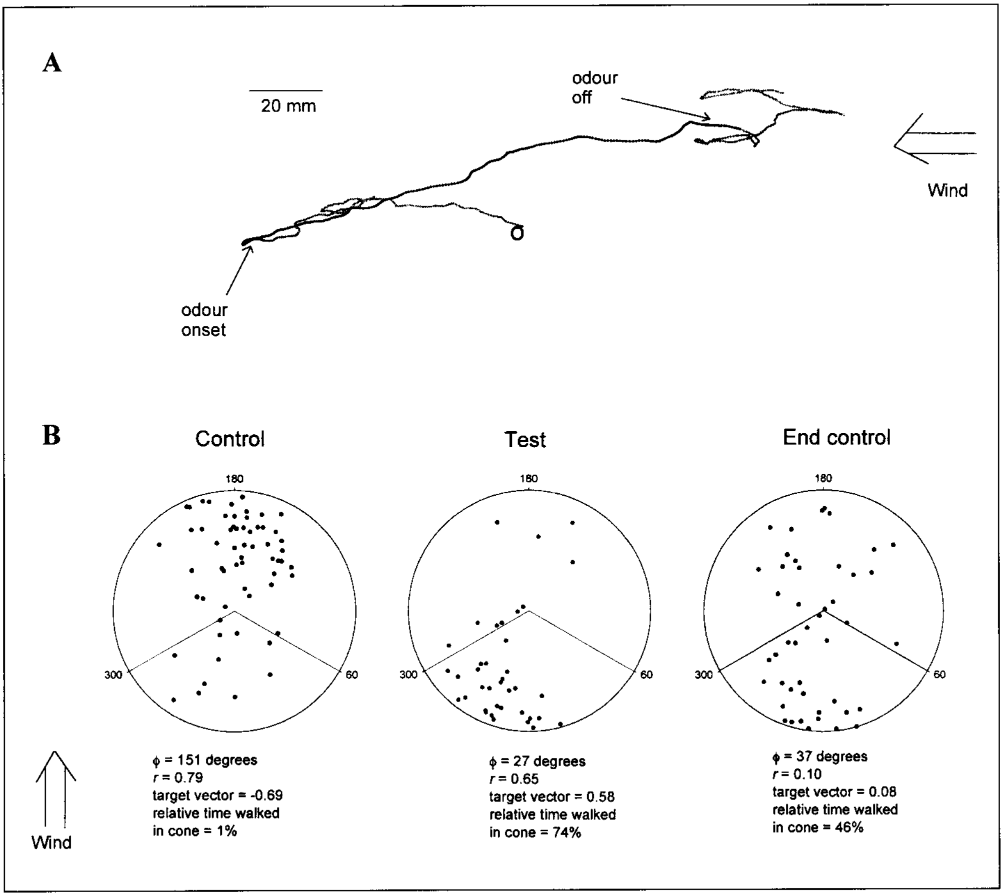

Fig. 4 (*A*) Response of an adult male *Amblyomma variegatum* on the servosphere to human breath diluted 25 times in the conditioned air stream flowing over the servosphere for one minute. The track commenced (o) with the tick walking downwind in the initial 1-minute control period. On delivery of breath (odour onset), the tick turned and undertook an upwind walk towards the source. In the 1-minute record of the walk after odour off (end control) the tick undertook a tortuous local search. (*B*) Polar plots of the distribution of 1-second vectors of the 3-minute track in A divided into control, test and end control periods. The distance from the origin represents the length of a vector and the perimeter of the circle a displacement of 30 mm. The mean direction ø, and the path straightness *r* (circular statistics, after BATSCHELET 1981) are indicated underneath the polar plots for each period. The degree of displacement upwind can be estimated by the target vector (*r*.cos ø, ranging from –1 downwind to +1 upwind) or, alternatively, by the relative time spent walking in an upwind cone (here 60° either side of due upwind).

mines respond to short-chain fatty acids and aldehydes such as methylpropanoic acid and nonanal (Fig. 5; GUERENSTEIN and GUERIN 2001).

An ammonia receptor predominates in grooved-peg sensilla on triatomine antennae, and this product on its own attracts triatomines (Tab. 1). Ammonia is almost as ubiquitous as CO_2 around vertebrates, occurring in breath, body odour and over urine (TANEJA and GUERIN 1997). Ammonia is also released from wetted triatomine faeces, and triatomine faeces are attractive to the bugs (TANEJA and GUERIN 1997). It would appear that the triatomines make parsimonious use of ammonia, as an odour cue for host finding and in the detection of well frequented refuges by cogeners where regularly deposited faeces cause ammonia to be released.

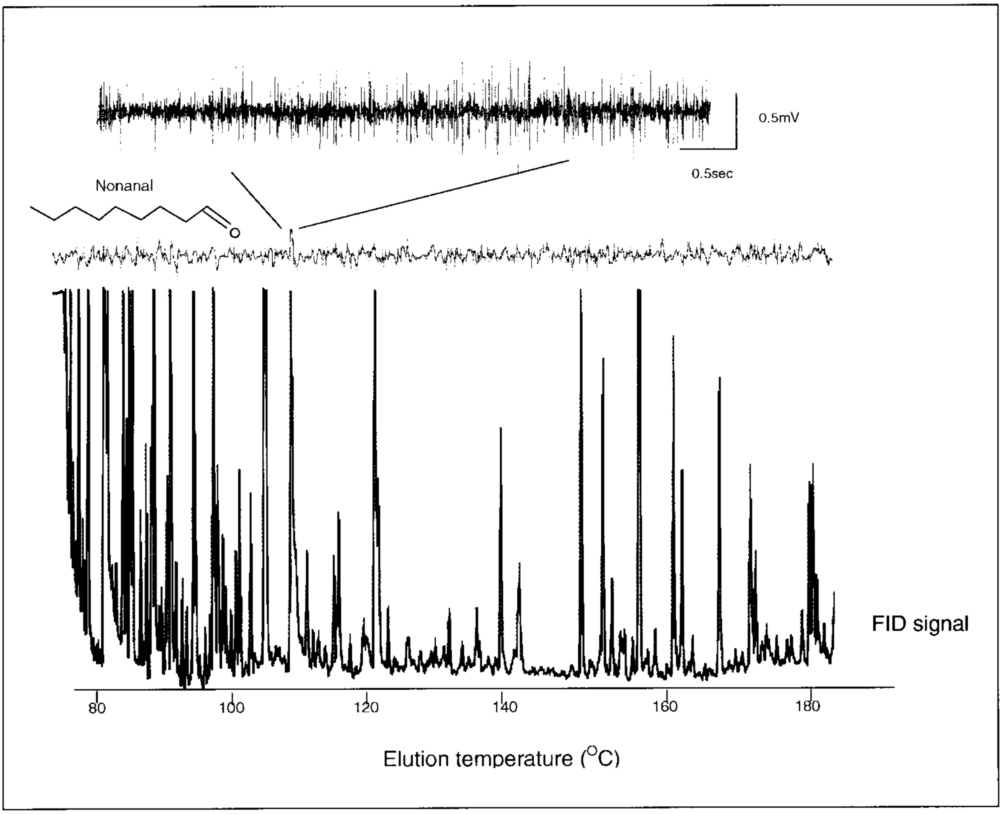

Fig. 5 Analysis of chicken-feather odour as collected on a porous polymer by gas chromatography coupled electrophysiology recording from olfactory receptors in a wall-pore sensillum on the antenna of a 5th instar *Triatoma infestans* nymph. The *lower trace* (blue) is the flame ionization detector (FID) response of the gas chromatograph, and the *middle trace* (red) is the summed frequency of all firing cells in the sensillum (frequency to voltage converted signal). The *upper trace* (black) is the actual action potential train generated during elution from the chromatograph of the biologically active peak in chicken-feather odour. The active peak (ca. 25 ng) was identified as nonanal since the electrophysiological response was the same to a similar amount of synthetic nonanal that eluted from the gas chromatographic column at the same retention time as the active product in the extract. Note the selective response of the receptors (a total of between 21 and 41 occur in such wall-pore sensilla) to just one of many products in the chicken-feather odour bouquet.

Tab. 1 Response of adult *Triatoma infestans* ($n = 11$) on the servosphere to 1 ppb ammonia presented in the air flowing over them. Each repetition consisted of a 2-minute control period followed by 2-minute test period with 1 ppb ammonia. Upwind displacement in response to ammonia was calculated by the difference from control of the %time spent walking in the upwind cone 60° either side of upwind, the %distance walked in that cone, and target vector (see legend to Fig. 4). Values presented are medians, and significance levels were established with the two-tailed Wilcoxon signed rank test for paired replicates ([1] $p < 0.01$ %).

	% Time in the upwind cone	% Distance walked in the upwind cone	Target vector
Control	2.1	1.98	–0.7
Test	52.05[1]	45.54[1]	0.43[1]

Lutzomyia longipalpis is the sandfly vector of visceral leishmaniasis in South America. Like triatomines, these insects occupy peridomestic niches, ovipositing on organic matter in the crevices of chicken coops and cow pens. Antennal segments 3 to 15 (total of 18 segments) bear a pair of ascoid sensilla set proximally but running almost the same length as each antennal segment (ca. 35 μm long). These large wall-pore olfactory sensilla house 8 sensory cells with ramified dendrites (ca. 30). By analysing rabbit and chicken faecal volatiles by gas chromatography linked to electrophysiological recordings from ascoid sensilla (see Fig. 5 for method) we identified hexanal and 2-methyl-2-butanol as chemostimulants, and a mixture of these two products caused targeted oviposition near the source in a cage bioassay (DOUGHERTY et al. 1995). This ascoid sensillum was also shown to house a receptor for a homosesquiterpene attractant for female *L. longipalpis*, secreted by tergal glands of the male during lek formation near hosts (DOUGHERTY et al. 1995). Volatiles from the scent glands of the fox, the principal sylvatic zoonotic reservoir of leishmaniasis in South America, are also perceived by *L. longipalpis* ascoid sensillum receptors (DOUGHERTY et al. 1999). Here again the short-chain fatty acids and aldehydes figure among the chemostimulants.

4. Signalling Suitable Hosts to Conspecifics

Host finding can be facilitated through prior occupation on or near a host by conspecifics signalling their presence there through the production of species-specific volatile attractants or pheromones from exocrine glands. A case in point is the production of a homosesquiterpene by tergal glands in male sandflies that form leks near hosts. The attractant serves to attract females to the leks, thus facilitating both encounter between the sexes for reproduction and subsequent feeding on the host (HAMILTON et al. 1994). A similar phenomenon is known for mosquitoes where presence of female mosquitoes on the host significantly enhances the host seeking responses of conspecifics (AHMADI and MCCLELLAND 1985).

Male bont ticks employ a similar strategy to lure conspecifics to a suitable host. In the case of *A. variegatum*, once pioneer males have found a host and successfully attached, dermal glands type 2 develop and release the volatile aggregation-attachment pheromone (Fig. 6; SCHÖNI et al. 1984, DIEHL et al. 1991). This mixture of volatiles, which includes *o*-nitrophenol and methyl salicylate as key components, serves to attract conspecifics (Fig. 7; MCMAHON and GUERIN 2000). Hosts with successfully attached pheromone-producing males attract more ticks (NORVAL et al. 1989). These secondary colonizers are lured to the host through the combined effects of attractive host odours and the pheromone (Fig. 6; MCMAHON and GUERIN 2000). The attached males clasp the subsequently arriving females so efficiently that host-attached venter to venter mating-pairs are formed in dense clusters at the preferred attachment sites (Fig. 8).

Numerous advantages accrue from such a host-colonization strategy in bont ticks. As it is adult *A. variegatum* males that undertake prospecting for hosts, the egg-laying females are preserved from the vagaries of the host-finding endeavour that includes exposition to predators once out of resting sites on the ground, and partial engorgement on a tick-resistant host. Males are better equipped for host prospecting. The male idiosoma is different from that of females in that males possess a coloured scutum whereas females do not have such a sclerotised covering on the posterior part of the idiosoma (Fig. 8). The coloured male scutum could serve to reinforce through association (Rowe and Guilford 1996) the repulsive message transmitted to would-be predators *via* defensive secretions from dermal glands (Pavis et al. 1994). Furthermore, acquired resistance to ticks is well established in vertebrates and ticks attached

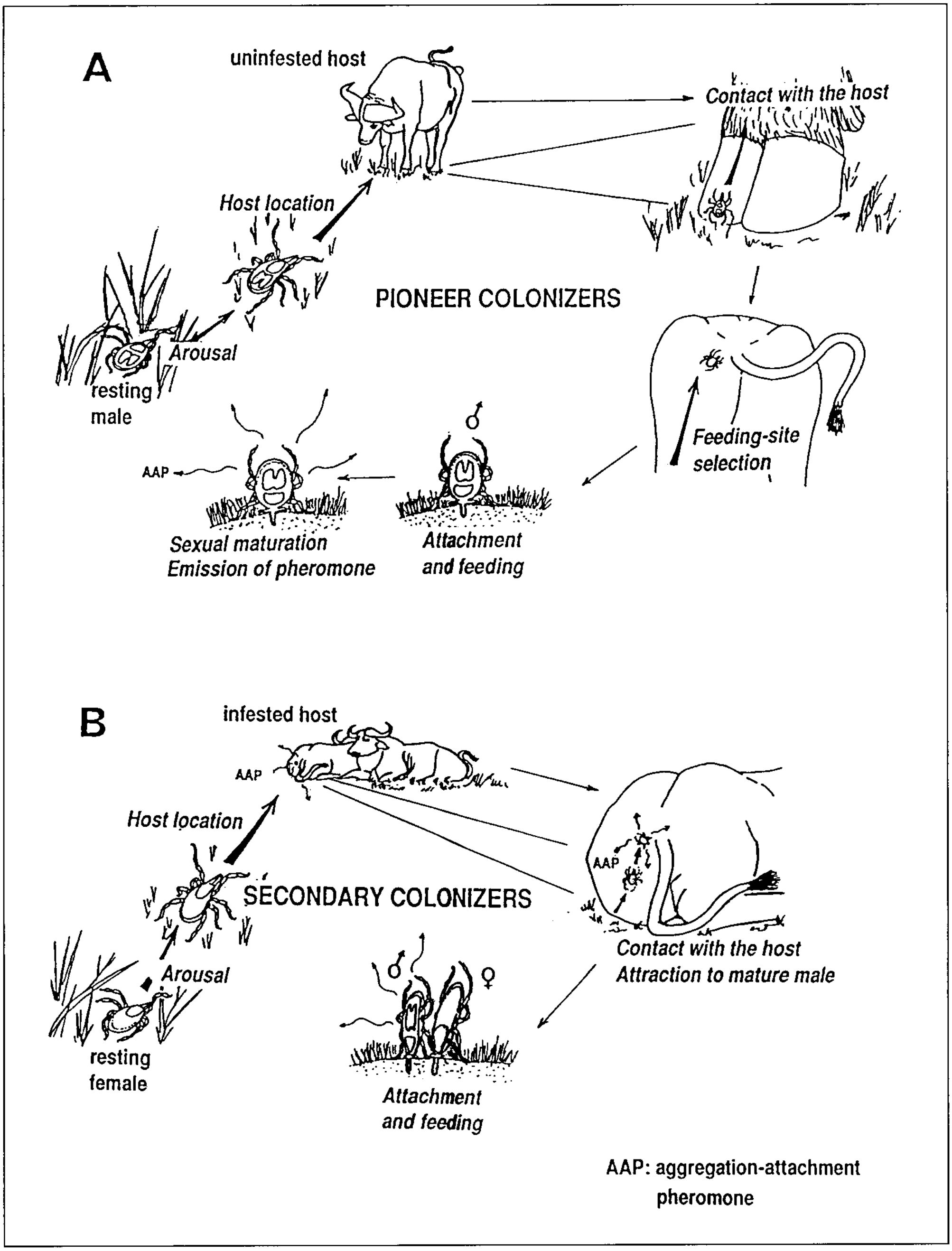

Fig. 6 In the tropical bont tick, *Amblyomma variegatum*, pioneer males (*A*) first find a suitable host and, after attachment, emit an aggregation-attachment pheromone that (*B*) attracts secondary colonizers including females to form host-attached mating pairs at the preferred feeding sites (see Fig. 8).

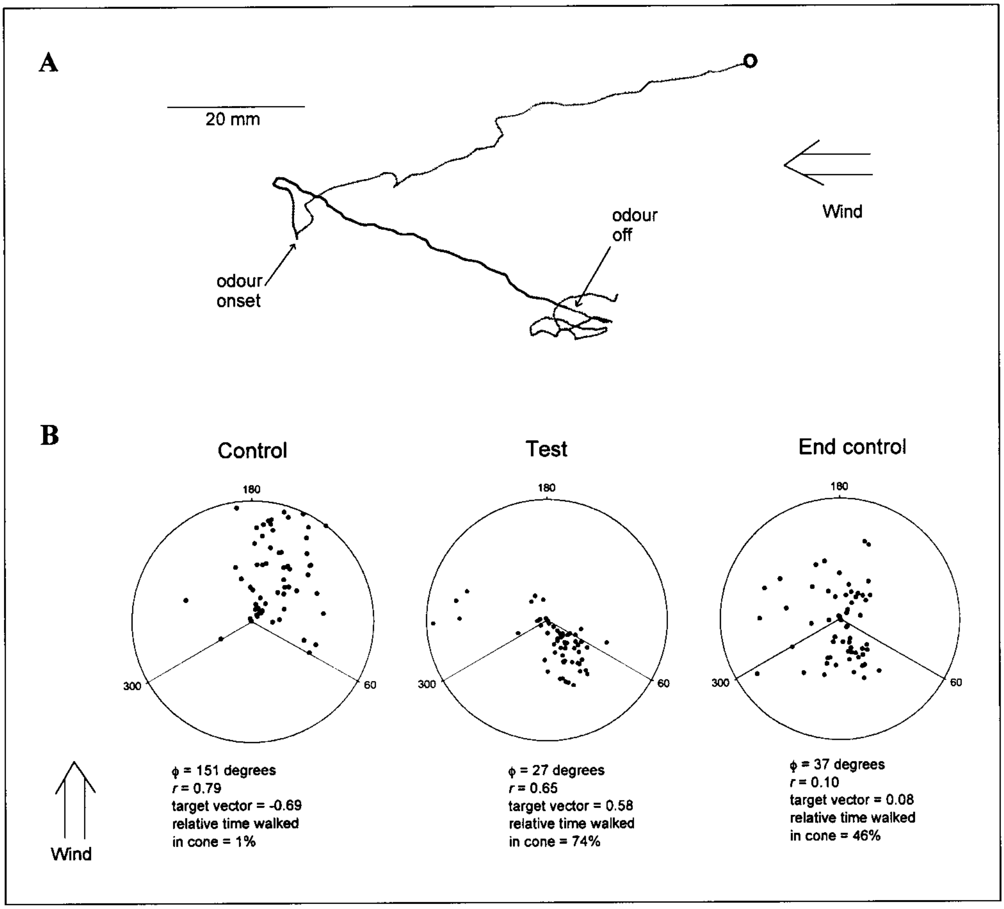

Fig. 7 (*A*) Response of an adult male *Amblyomma variegatum* on the servosphere to its aggregation-attachment pheromone (vapours from a 1 : 1 mixture of *o*-nitrophenol and methyl salicylate; source dose 10 ng each) presented in the conditioned air stream flowing over the servosphere for one minute. The track commenced (o) with the tick walking downwind in the initial 1-minute control period. On delivery of the aggregation-attachment pheromone vapour (odour onset), the tick turned and undertook an upwind walk towards the source. In the 1-minute record of the walk after odour off (end control) the tick undertook a tortuous local search. (*B*) Polar plots of the distribution of 1-second vectors of the 3-minute track in (*A*) divided into control, test and end control periods. For further explanation, see legend to Fig. 4 *B*.

to tolerant hosts fail to obtain an optimal blood-meal (LÖSEL et al. 1992, 1993). Since ingestion of host fluid triggers cuticle synthesis in females to accommodate the blood-meal, this sex would run the risk of acquiring a permanent weight gain on an unsuitable host that would handicap mobility during subsequent host prospecting. As males possess an inflexible scutum they are prohibited from such a weight gain and so may risk prospective attachment to an unsuitable host. In addition, female ticks will not feed to repletion without being fertilized (OLIVER 1989), so obtaining sperm is at a premium. But female attraction to male aggregations on the host increases the probability of being mated. Consequent to this aggregation, co-feeding at preferred feeding sites could serve as a further advantage for *A. variegatum* in overcoming host defence reactions, as has been shown for *Rhipicephalus appendiculatus*

Fig. 8 *Amblyomma variegatum* mating pairs on a rabbit. Males have a coloured scutum, and the three females on the photo are at various stages of engorgement.

(Wang et al. 1998). It is of interest that a similar strategy of host plant colonization is known for bark beetles, where pioneering scolytid invaders of pine trees release species specific attractants that serve to recruit conspecifics to the invasion site so as to overcome the host-tree mobilization of defensive phloem flow (Vité and Francke 1976). A further advantage for bont ticks lies in the nature of the aggregation-attachment attractants produced by the dermal glands of *A. variegatum* males. As in the bark beetle model, the pheromone released by *A. variegatum* males includes a host volatile, *o*-nitrophenol. When this product is presented alone to *A. variegatum* adults walking on the servosphere it is not a strong attractant, but is attractive when combined with host hair – a substrate that is unattractive on its own (McMahon and Guerin 2000). So this bont tick species may be enhancing the release of a critical host volatile in its aggregation-attachment pheromone to render suitable hosts more apparent to conspecifics (Steullet 1993). Finally, even after female drop-off, males stay attached to the host, ensuring the continued attraction of the resource to other conspecifics.

5. Contact-Mediated Host-Attachment Stimuli

There are only a few detailed studies available on attachment stimuli for blood-feeding arthropods. A combination of uric acid and heat was shown to induce probing by the tsetse fly *Glossina fuscipes fuscipes* (van Naters et al. 1998) and chemosensory hairs on tsetse fly legs possess chemoreceptors for different amino acids present in human sweat (van Naters and den Otter 1998). Among acarids, work on the red

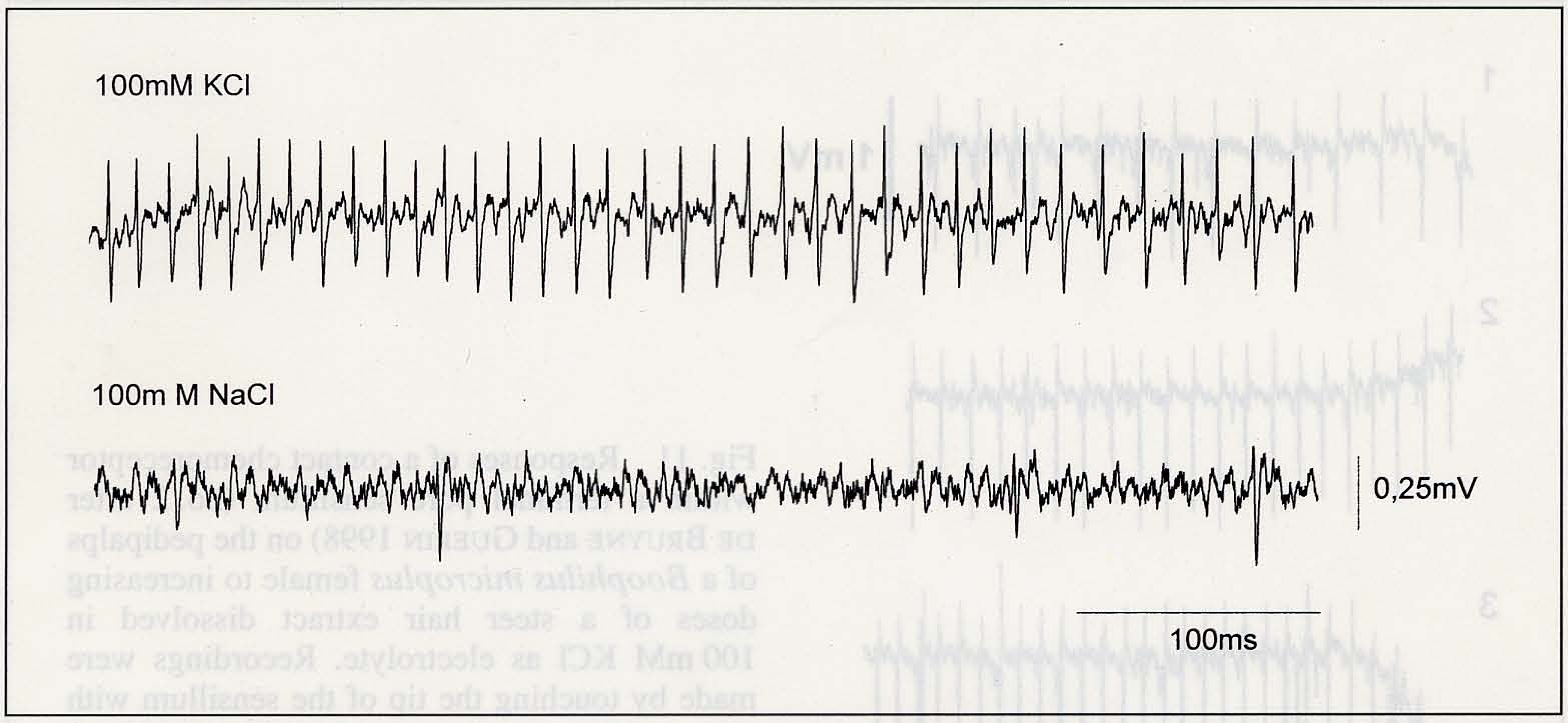

Fig. 9 An electrophysiological response (train of action potentials) to 100 mM KCl but not to 100 mM NaCl by a receptor in a terminal-pore contact chemosensillum on a pedipalp of *Ixodes ricinus*. The recordings were made by touching the terminal-pore area of the sensillum with the tip (10 µm diameter) of a glass capillary filled with either electrolyte and linked *via* a silver wire to an amplifier. For further information see legend to Fig. 11.

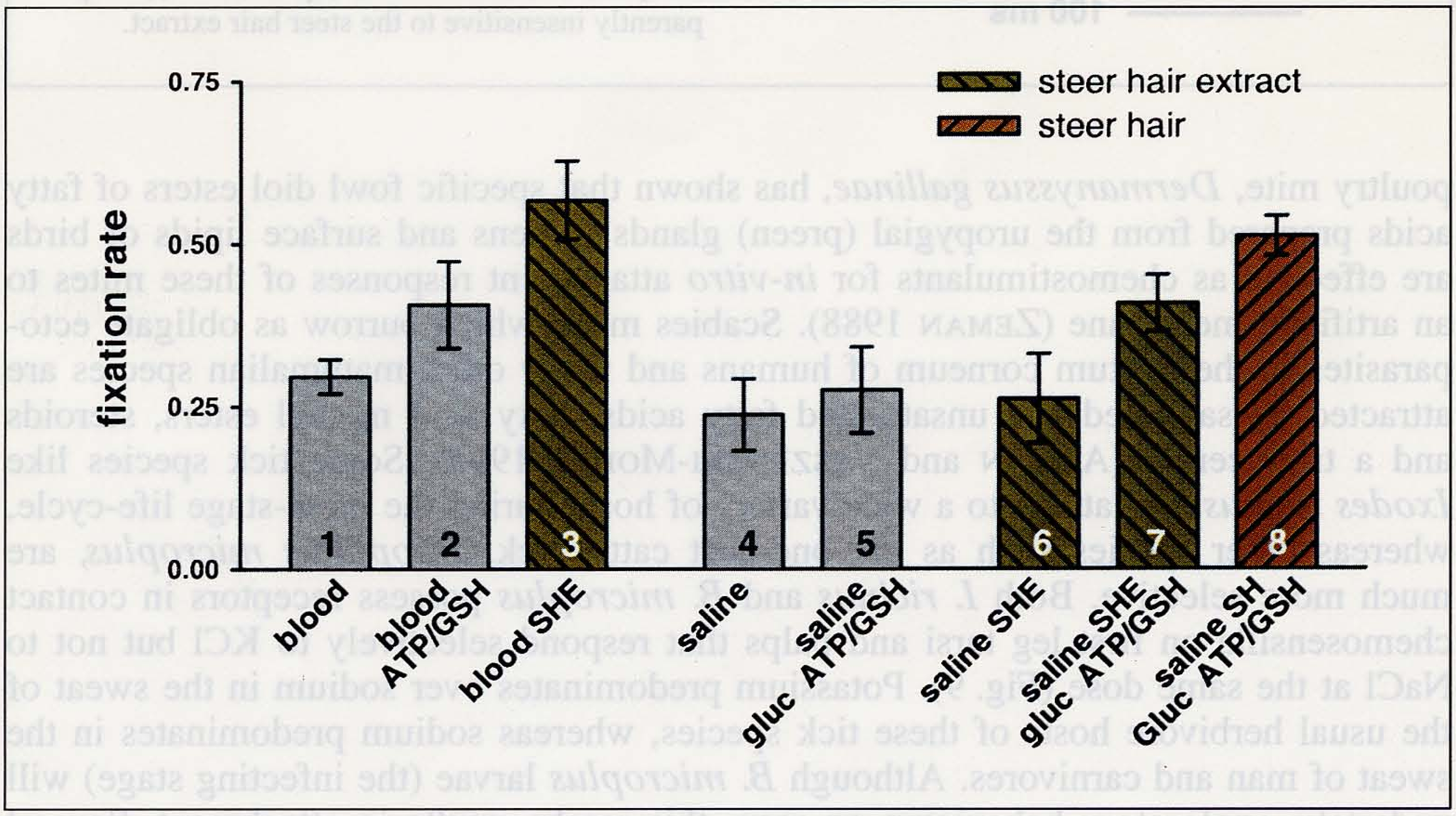

Fig. 10 *In-vitro* attachment rates of *Amblyomma hebraeum* nymphs to a membrane stretched over a reservoir containing either blood or saline at 37 °C (Kuhnert et al. 1995). The membrane or the fluid beneath it were treated with different stimuli 1, bovine blood (n = 10), 2, bovine blood with 3 mM each of ATP and glutathion added (ATP/GSH, n = 8), 3, bovine blood covered with a membrane treated with 20 µg/cm^2 steer hair extract (SHE, olive, n = 9), 4, 0.9 % saline (n = 9), 5, saline with 3 mM each of ATP/GSH and 2 mg/ml glucose added (n = 10), 6, saline covered with a membrane treated with 20 µg/cm^2 steer hair extract (n = 10), 7, as 6 but with 3 mM ATP/GSH and 2 mg/ml glucose added, (n = 9), 8, as 7, but covered by a membrane treated with steer hair (SH, brown) rather than the steer hair extract (n = 7), n = number of the feeding units each with ca. 35 nymphs. The error bars provide a measure of the significance level ($p = < 0.05$; Elsner and Knutti 1991).

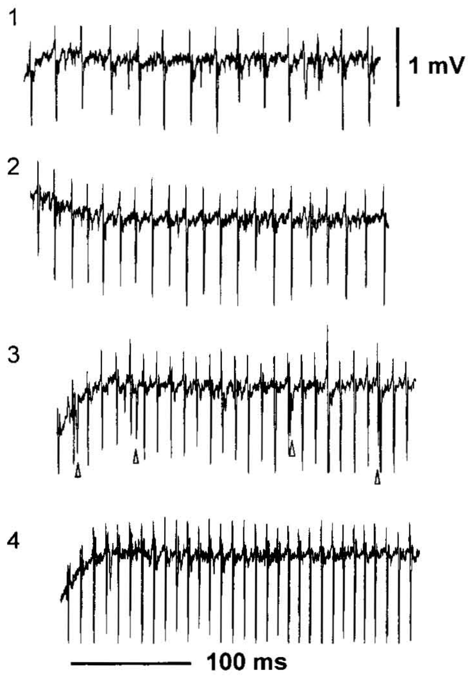

Fig. 11 Responses of a contact chemoreceptor within a terminal pore sensillum (no. 2 after DE BRUYNE and GUERIN 1998) on the pedipalps of a *Boophilus microplus* female to increasing doses of a steer hair extract dissolved in 100 mM KCl as electrolyte. Recordings were made by touching the tip of the sensillum with the stimulus contained in the open tip (5 µm diameter) of a drawn-out glass capillary linked *via* a silver wire in the electrolyte to a high impedance preamplifier; the reference electrode (ground) was placed in the tick's body. The firing rate of the receptor cell rises with increasing dose of the steer hair extract 1, 5 pg lipids/ml; 2, 0,5 ng/ml; 3, 5 ng/ml; 4, 5 µg/ml. A second receptor, visible in trace 3 (open arrows), is apparently insensitive to the steer hair extract.

poultry mite, *Dermanyssus gallinae*, has shown that specific fowl diol esters of fatty acids prepared from the uropygial (preen) glands of hens and surface lipids of birds are effective as chemostimulants for *in-vitro* attachment responses of these mites to an artificial membrane (ZEMAN 1988). Scabies mites which burrow as obligate ectoparasites in the stratum corneum of humans and many other mammalian species are attracted by saturated and unsaturated fatty acids, fatty acid methyl esters, steroids and a triglyceride (ARLIAN and VYSZENSKI-MOHER 1995). Some tick species like *Ixodes ricinus* will attach to a wide variety of hosts during the three-stage life-cycle, whereas other species such as the one-host cattle tick, *Boophilus microplus,* are much more selective. Both *I. ricinus* and *B. microplus* possess receptors in contact chemosensilla on first leg tarsi and palps that respond selectively to KCl but not to NaCl at the same dose (Fig. 9). Potassium predominates over sodium in the sweat of the usual herbivore hosts of these tick species, whereas sodium predominates in the sweat of man and carnivores. Although *B. microplus* larvae (the infecting stage) will undertake exploratory behaviours on man, this rarely results in attachment. Several tick species show preferences for attachment on particular regions of the host's body, such as host ears for *R. appendiculatus*. It has long been suggested that regional differences in the stimuli provided might influence these predilection behaviours.

Much of our knowledge in the domain of contact-mediated host-attachment responses of ticks to skin lipids has been gathered during development of an *in-vitro* feeding system for hard ticks (KUHNERT et al. 1995, KUHNERT 1996). When an organic solvent extract of steer hair was applied to an artificial membrane it caused a significantly higher number of ticks to attach and feed from a blood reservoir beneath

(Fig. 10). Macro-video has permitted us to observe that ticks regularly sample the substrate on which they walk with the palps, the tips of which carry numerous terminal-pore contact chemosensilla. The firing rate of a chemoreceptor in one of these sensilla in *B. microplus* increases with increasing dose of a steer hair extract applied to the tip of the recording electrode (Fig. 11). *In-vitro,* ticks confine themselves to zones of semipermeable membranes rendered hydrophobic by treatment with a host lipid extract, clearly avoiding any contact with a surrounding wet membrane surface (KRÖBER and GUERIN 1999). Avoidance of wet surfaces on the host ensures that ticks do not attach to feed from highly innervated surfaces such as lips and mouth cavity, nose, eyes or anus. Because of the degree of disturbance it would cause to the host by attachment at such sites, it is evidently of survival value for the tick to avoid such places on the host.

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