

## HISTAMINE-INDUCED MODULATION OF OLFACTORY RECEPTOR NEURONES IN TWO SPECIES OF LOBSTER, *PANULIRUS ARGUS* AND *HOMARUS AMERICANUS*

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### Summary

In two species of lobster, application of the biogenic amine, histamine (HA), to the soma of olfactory receptor cells suppressed both spontaneous and odour-evoked activity, as shown by electrophysiological recording from single cells. The action of HA was graded, reversible, specific to HA, and had a threshold between 0.1 and 1  $\mu\text{mol l}^{-1}$ . HA increased the conductance of the membrane, primarily to chloride ions. The vertebrate HA receptor antagonist, cimetidine, and the nicotinic receptor antagonist, *d*-tubocurarine, but not other known vertebrate HA receptor antagonists, reversibly blocked the action of HA. These results suggest that a histaminergic mechanism modulates stimulus–response coupling in lobster olfactory receptor cells and potentially implicate a novel HA receptor, pharmacologically similar to the one recently described in the visual system of flies.

### Introduction

There is growing evidence that sensory systems are subject to peripheral as well as central modulation (e.g. Flock & Russell, 1973; Art *et al.* 1982; Pollock *et al.* 1985; Siegelbaum *et al.* 1986; Cuello, 1987). Although the concept of peripheral modulation is probably least well established for the chemical senses, there are now hints that olfaction can be modulated peripherally in insects (Davis & Takahashi, 1980) and vertebrates (Bouvet *et al.* 1987; Yoshioka *et al.* 1987). To investigate peripheral modulation in olfaction, we have been surveying the effect of known neuroactive compounds on crustacean olfactory receptor cells, which have been the focus of recent research in peripheral olfactory organization (e.g. Ache & Derby, 1985). Potential neuromodulators for crustacean olfactory receptor cells include a number of substances that have been shown to act on

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crustacean neurones (for a review see Kravitz, 1988), including some having actions on primary sensory neurones (Hagiwara *et al.* 1960; Jansen *et al.* 1971; Pasztor & Bush, 1987). We now report that histamine (HA) modulates the sensitivity of lobster olfactory receptor cells.

HA is a compound of long-standing neurobiological importance (Schwartz *et al.* 1979; Prell & Green, 1986) that has been associated with diverse effects in many species (Reite, 1972; Kehoe & Marder, 1976). It is a putative neurotransmitter in the mammalian central nervous system (Schwartz *et al.* 1980), where it has a modulatory function (Haas, 1984). Recently, there has also been growing interest in HA as a putative neurotransmitter in invertebrates and it has now been implicated as a neurotransmitter in molluscs (Weinreich, 1977; McCaman & Weinreich, 1982, 1985), crustaceans (Claiborne & Selverston, 1984; Stuart & Callaway, 1988) and insects (Hardie, 1987, 1988; Simmons & Hardie, 1988). The functional importance of HA-mediated effects, however, is not always clear (Kerkut *et al.* 1968; Carpenter & Gaubatz, 1975; Gotow *et al.* 1980; Ku & Takeuchi, 1983), and further work on this interesting molecule is in order.

## Materials and methods

### *Animals*

Adult, intermoult specimens of the spiny lobster *Panulirus argus* were collected in the Florida Keys and maintained in flowing sea water at 20–23°C. Similar specimens of the clawed lobster *Homarus americanus* were obtained from commercial suppliers in Woods Hole, MA, and maintained in flowing sea water at 13–17°C. The spiny lobster was used for all experiments unless noted otherwise.

### *Preparation*

The lateral filament of the antennule (the olfactory organ, Fig. 1) was excised and cut into pieces approximately 1 mm long. These pieces were halved longitudinally and the halves containing the hair-like aesthetasc (olfactory) sensilla were treated with enzymes ( $\text{l-cysteine-activated papain}$ : Sigma Type IV,  $0.25 \text{ mg ml}^{-1}$  saline, 25 min; followed by  $\text{trypsin}$ : Sigma Type IX,  $0.2 \text{ mg ml}^{-1}$   $\text{Ca}^{2+}$ -free saline, 25 min). After rinsing three times with lobster saline (Mulloney & Selverston, 1974), single sections were placed in a silicone elastomer recording chamber (Fig. 1). The preparation fitted tightly into a slit between an upper saline bath and a tube below. The hairs projected into the tube, and the somata of the receptor neurones were exposed in the bath. The tube was continuously perfused ( $4 \text{ ml min}^{-1}$ ) with saline. Replicate,  $20 \mu\text{l}$  samples of an odorant injected into this flow *via* a rotary sample injection valve (Rheodyne 5020) arrived at the hairs approximately 2 s after injection with no concomitant mechanical stimulation. The anatomy of the olfactory sensilla (Grünert & Ache, 1988) is such that stimulation of the hairs selectively stimulates the outer dendritic segments of the olfactory receptor neurones. The solution bathing the somata could be changed independently of that bathing the outer dendritic segments. Alternatively, drugs were

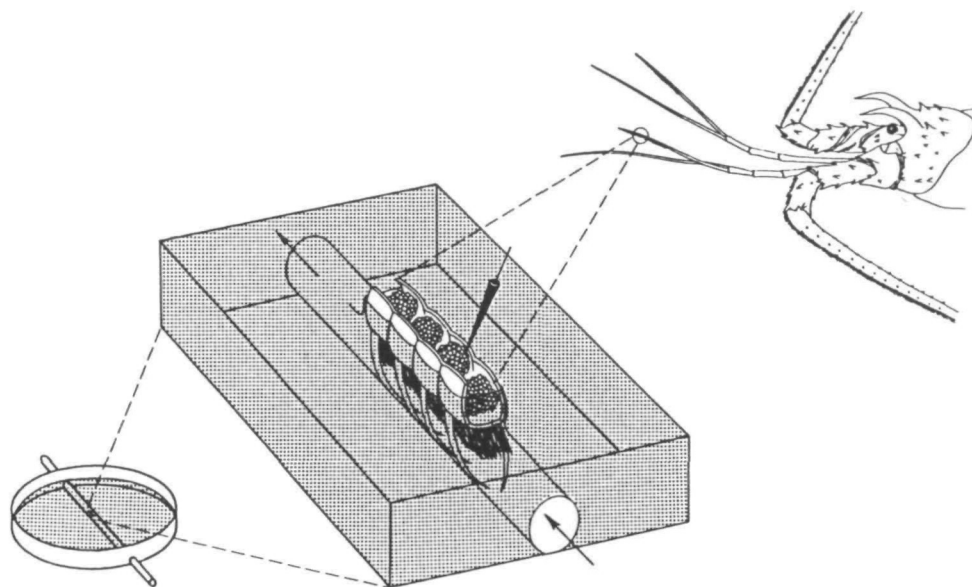


Fig. 1. Diagram of *in situ* preparation for recording from the receptor cells of lobster olfactory sensilla. The hair-like olfactory sensilla (aesthetascs) are borne as an orderly tuft on the lateral branch of each of the paired antennules. See text for details.

directly applied to the somata either by pressure ejecting them from a micropipette positioned near the recording site or from a hand-held digital microlitre pipette.

For some experiments, somata were removed from the pieces by a single trituration step, a process which stripped off the majority of the dendrites, and dispersed into a saline-filled 35 ml culture dish. The somata of olfactory receptor cells were identified by their spherical shape and their diameter of 10–20  $\mu\text{m}$  (Grünert & Ache, 1988). All work was done at room temperature (approx. 22°C).

### Recording

Extracellular recording was achieved by applying saline-filled patch electrodes to the soma and using suction to achieve seals of sufficient resistance to resolve action potentials. These electrodes were pulled from borosilicate glass (Boralex), firepolished to a bubble number of 3.5–4.5 (Mittman *et al.* 1987), and filled with saline. Signals were amplified with a conventional high-impedance, capacity-coupled amplifier (Grass P511) and displayed on an analogue storage oscilloscope (Tektronix 5111). Action potentials were converted into TTL pulses with a Schmitt trigger and the resulting intervals analysed by computer and stored on floppy disk.

Intracellular recordings were obtained from either intact cells (current-clamp) or isolated somata (voltage-clamp) by applying patch electrodes to the soma in the whole-cell configuration (Hamill *et al.* 1981). Intracellular electrodes were pulled from the same glass as that used for extracellular electrodes, but were unpolished

(4.8–5.2 bubble number) and filled with patch solution. Silicone elastomer (Sylgard 184) was applied to the neck of pipettes used for voltage-clamp recording. After forming a seal and compensating the electrode capacitance, the whole-cell configuration was achieved using suction to break through the cell membrane and the series resistance was compensated qualitatively. Signals were amplified with a conventional patch amplifier (Dagan 8900), filtered at a corner frequency of 10 kHz and stored on video tape (Bezanilla, 1985). Data were analysed off-line from records displayed either on a chart recorder (Gould 220) or a digital oscilloscope (Nicolet 4094).

Intact cells were verified as functional chemoreceptors by their ability to respond in a graded manner to successive dilutions of a standard odorant (see below), but not to an identically handled saline blank.

#### *Solutions and chemicals*

The composition of lobster saline was (in  $\text{mmol l}^{-1}$ ): NaCl, 458.0; KCl, 13.4;  $\text{MgCl}_2$ , 9.8;  $\text{CaCl}_2$ , 13.6;  $\text{Na}_2\text{SO}_4$ , 14.0; Hepes, 10.0; glucose, 2.0; pH 7.4.  $\text{K}^+$ -free saline was prepared by substituting  $\text{K}^+$  with  $\text{Na}^+$ .  $\text{Na}^+$ -free saline was prepared by substituting  $\text{Na}^+$  with tetramethylammonium chloride (TMA-Cl). Normal patch solution consisted of (in  $\text{mmol l}^{-1}$ ): KCl, 26.0; potassium acetate, 180.0; EGTA, 11.0; Hepes, 10.0;  $\text{CaCl}_2$ , 1.0; glucose, 696.0; pH 7.0. High-chloride patch solutions consisted of either (in  $\text{mmol l}^{-1}$ ): KCl, 140.0; EGTA, 11.0;  $\text{CaCl}_2$ , 1.0; Hepes, 10.0; glucose, 696.0; pH 7.0; or KCl, 200.0;  $\text{MgCl}_2$ , 2.0; EGTA, 11.0;  $\text{CaCl}_2$ , 1.0; Hepes, 10.0; glucose, 570.0; pH 7.0.

Drugs were prepared on the day of use by dissolving them in the saline. Acetylcholine, carnosine, cimetidine, dopamine, DL-glutamate, DL-octopamine, *d*-tubocurarine,  $\gamma$ -aminobutyric acid, histamine, histidine, 1-methyl-histamine, L-aspartate, proctolin, pyrilamine and serotonin were purchased from Sigma. Burimamide, dimaprit and impromidine were gifts from Dr D. Weinreich.

The standard odorant was prepared as a stock solution by dissolving 2 g of TetraMarin (TET), a commercially prepared, dry flake food for marine fish (TetraWerke, Melle, FRG) consisting of both plant and animal material, in 60 ml of saline, adjusting to pH 7.4 and filtering the resulting extract to remove particulate material. The filtrate was frozen in 5 ml samples that were thawed and serially diluted in saline on the day of use.

## **Results**

Selectively applying HA to the *soma* suppressed the spontaneous and the odour-evoked activity in 86 of the 95 olfactory receptor cells tested in this study (Fig. 2). Since HA is also an effective odorant for these cells (B. W. Ache, B. R. Johnson & E. Clark, unpublished data), we tested if cells suppressed by somatic application of HA were also sensitive to HA applied as an odorant (i.e. dendritic application). There was no strict relationship between the somatic and dendritic actions of HA. In 35 of 53 cells on which both actions were tested, HA was either excitatory or

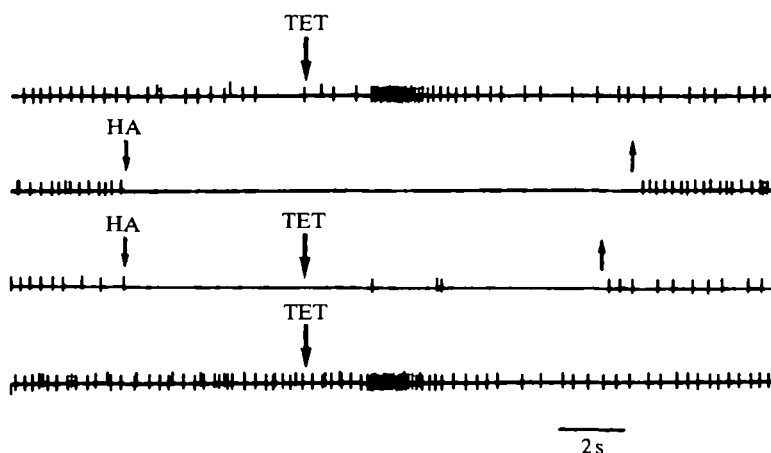


Fig. 2. Extracellular recordings from a lobster olfactory receptor cell. Top trace, excitatory response to dendritic application (arrow) of 1:1000 dilution of the standard odorant, TetraMarin (TET). The arrow denotes the time of injection, 2s prior to arrival of the odorant at the preparation. Second trace, spontaneous activity suppressed by somatic application (downward arrow) of  $10^{-5} \text{ mol l}^{-1}$  histamine (HA). Block relieved by saline rinse (upward arrow). HA applied by replacing the bath solution. Third trace, both spontaneous activity and the excitatory response to dendritic application of TET (large arrow) are suppressed by somatic application of  $10^{-5} \text{ mol l}^{-1}$  HA (small downward arrow). The block is relieved by a saline rinse (small upward arrow). Last trace, recovery of the excitatory response to dendritic application (arrow) of TET.

had no effect as an odorant. HA also excited two cells as an odorant on which it had no effect when applied somatically. Eighteen of the 53 cells suppressed by somatic application of HA, however, were also suppressed by dendritic application of the drug. Although it is unlikely that HA leaked between the two compartments, we could not exclude this possibility with absolute certainty and therefore report data on the somatic effects of HA only from cells shown to be excited or unstimulated by HA applied as an odorant or from cells in which focal presentation of HA virtually eliminated the possibility of access to the dendrites.

The suppression of spiking by HA was immediate (Fig. 2) and typically persisted for the duration of exposure to the drug, the longest interval tested being 10 min ( $N=8$ ). In a few instances of prolonged exposure, however, the cells recovered their spontaneous activity after about 3 min, in spite of the persistence of the drug. Recovery was also rapid (Fig. 2); the cells recovered either immediately or within several seconds of washout of HA. Increasing the concentration of odorant used to evoke a response overcame the HA-induced suppression of excitability (data not shown,  $N=8$ ). The apparent threshold of the effect, measured by replacing the entire bath with solutions of different concentrations of HA to obtain an accurate measure of concentration, was between  $10^{-7} \text{ mol l}^{-1}$  (one out of seven cells suppressed) and  $10^{-6} \text{ mol l}^{-1}$  (five out of 13

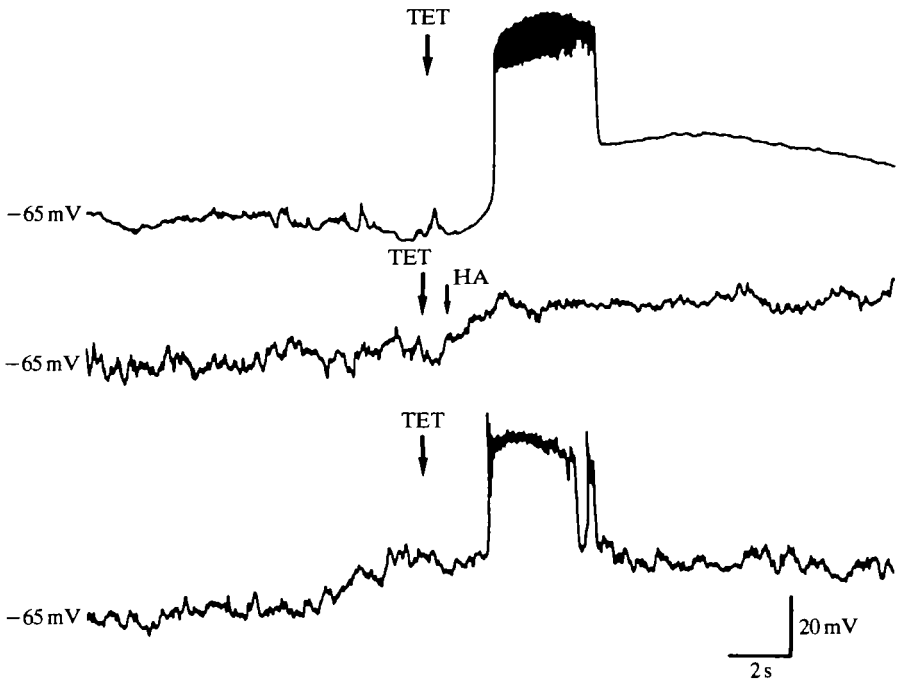


Fig. 3. Whole-cell current-clamp recordings from a lobster olfactory receptor cell (membrane potential set to  $-65$  mV, normal patch solution:  $E_{Cl} = -66$  mV). Top trace, suprathreshold depolarization evoked by dendritic application (arrow) of 1:10 dilution of the standard odorant (TET). Middle trace, pressure ejecting  $10^{-2}$  mol l $^{-1}$  HA onto the soma (500 ms, small arrow) reduced the effect of dendritic application of TET (large arrow). Bottom trace, the sensitivity to the dendritic application of TET (arrow) returned following removal of HA.

cells suppressed). The dose–response relationship was assessed intracellularly to obtain a graded measure of the effect (see below).

Somatically applied HA reversibly suppressed the amplitude of the odour-evoked receptor potential, recorded intracellularly (Fig. 3,  $N = 2$ ), which was consistent with the extracellular findings. The direct effect of HA under our usual recording conditions (normal patch solution, membrane potential set to  $-65$  mV) was to depolarize the cell transiently (data not shown,  $N = 4$ ). Voltage-clamp recordings from isolated somata indicated that HA induced a macroscopic current associated with an increase in membrane conductance (Fig. 4,  $N = 4$ ). The magnitude of the HA-induced current increased, then saturated, as a function of the duration of the pressure pulse used to eject  $10^{-3}$  mol l $^{-1}$  HA into the bath near the soma (Fig. 5,  $N = 5$ ). Given that the amount of HA delivered by the pipette is a linear function of the duration of pressure application (Van Dongen, 1984), the concentration of HA reaching the cell through the bulk phase could be expected to increase as a similar, although not necessarily identical, function of the pulse duration.

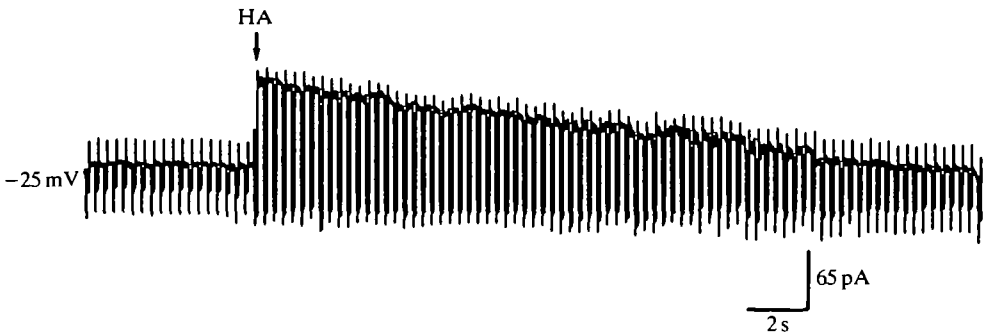


Fig. 4. Whole-cell voltage-clamp recording from the isolated soma of a lobster olfactory receptor cell (membrane potential held at  $-25$  mV, normal patch solution:  $E_{Cl} = -66$  mV). Pressure ejection of  $10^{-3}$  mol  $l^{-1}$  HA onto the soma (25 ms, arrow) induced an outward current associated with an increase in membrane conductance. Membrane conductance was monitored by injecting a 120 pA, 200 ms current pulse into the soma.

The HA-induced change in membrane potential was primarily the result of altered permeability to  $Cl^{-}$ . The response to somatically applied HA reversed polarity at  $-52$  mV (Fig. 6,  $N = 3$ ), slightly positive to the Nernst potential for chloride under our normal recording conditions ( $E_{Cl} = -66$  mV). Voltage-clamping isolated somata under altered ionic conditions further implicated  $Cl^{-}$  as the primary current-carrying ion. Increasing  $[Cl^{-}]_i$  to  $140$  mmol  $l^{-1}$  by using the appropriate high- $Cl^{-}$  patch solution ( $E_{Cl} = -31$  mV) shifted the reversal potential to a value more positive than that recorded under our normal recording conditions (Fig. 7A vs Fig. 6,  $N = 1$ ). A further increase in  $[Cl^{-}]_i$  to  $200$  mmol  $l^{-1}$  ( $E_{Cl} = -24$  mV) shifted the reversal potential to an even more positive value

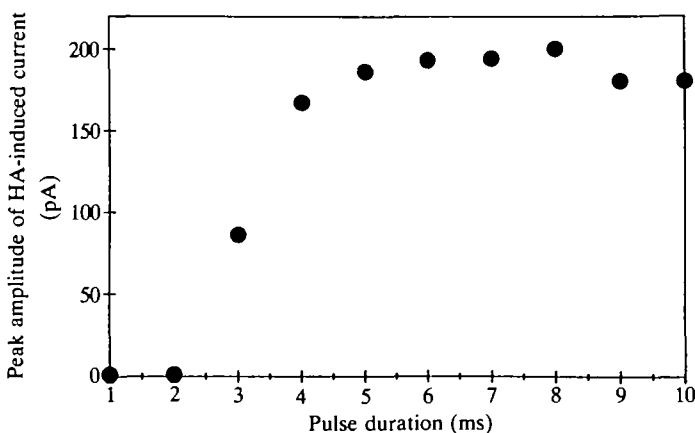


Fig. 5. Plot of the magnitude of the HA-induced current in an isolated soma of a lobster olfactory receptor cell under voltage-clamp as a function of the duration of the pressure pulse used to eject  $10^{-3}$  mol  $l^{-1}$  HA. The cell was held at  $0$  mV and normal patch solution was used ( $E_{Cl} = -66$  mV).

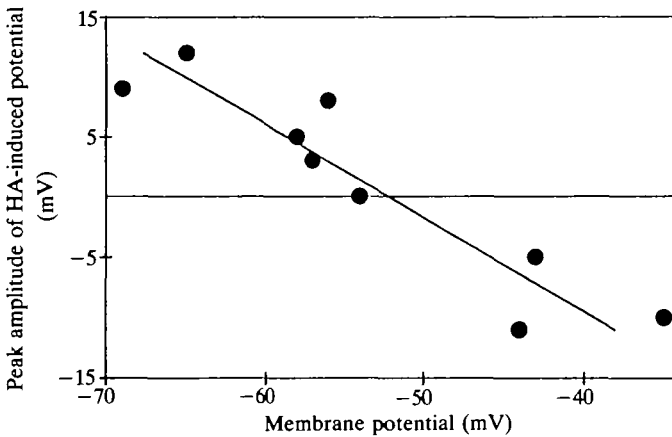


Fig. 6. Plot of the magnitude of the HA-induced polarization in lobster olfactory receptor cells as a function of membrane potential. The data shown are compiled from three different cells bathed in normal saline and perfused with normal patch solution ( $E_{Cl} = -66$  mV). In each instance  $10^{-2}$  mol l $^{-1}$  HA was pressure ejected onto the soma for 500 ms. The points regress to a straight line (plotted) with a correlation coefficient of 0.93. The mean reversal potential was  $-52 \pm 8$  mV (mean  $\pm$  s.d.).

(Fig. 7B,  $N = 4$ ). The observation that the current always reversed slightly positive to  $E_{Cl}$ , however, implicated secondary charge-carrying ion(s). Eliminating extracellular  $K^+$  elicited no marked effect on the reversal potential (Fig. 7C vs Fig. 7A,  $N = 4$ ), suggesting that  $K^+$  permeability was not altered by HA. Attempts to eliminate extracellular  $Na^+$  by substituting all the  $Na^+$  in normal saline with  $TMA^+$  gave variable results, preventing us from demonstrating a possible  $Na^+$  component. We cannot account for this discrepancy other than to note that we generally had difficulty obtaining repeatable data from isolated somata in the absence of  $Na^+$ . Single-channel evidence, however, indicates that the underlying chloride channel is partially permeable to  $Na^+$  (T. S. McClintock & B. W. Ache, in preparation).

The suppressive effect was specific to HA and could not be mimicked by any of the other compounds tested. Two catecholamines ( $10^{-4}$  mol l $^{-1}$  dopamine,  $N = 4$ ;  $10^{-6}$ – $10^{-4}$  mol l $^{-1}$  DL-octopamine,  $N = 20$ ), an indoleamine ( $10^{-4}$  mol l $^{-1}$  5-hydroxytryptamine,  $N = 5$ ), three amino acids ( $10^{-4}$  mol l $^{-1}$   $\gamma$ -aminobutyric acid,  $N = 3$ ;  $10^{-4}$  mol l $^{-1}$  L-aspartic acid,  $N = 4$ ;  $10^{-4}$  mol l $^{-1}$  DL-glutamic acid,  $N = 3$ ), three other imidazoleamines ( $10^{-4}$  mol l $^{-1}$  histidine,  $N = 4$ ;  $10^{-4}$  mol l $^{-1}$  L-carnosine,  $N = 3$ ;  $10^{-4}$  mol l $^{-1}$  1-methyl-histamine,  $N = 4$ ),  $10^{-4}$  mol l $^{-1}$  acetylcholine ( $N = 5$ ) and the peptide proctolin ( $10^{-4}$  mol l $^{-1}$ ,  $N = 4$ ) had no obvious effect on either spontaneous activity or the odour-evoked responses of the cells.

Preliminary pharmacological analysis of the effect showed that the vertebrate  $H_2$  antagonist, cimetidine, at  $10^{-3}$  mol l $^{-1}$  ( $N = 6$ ) and the nicotinic antagonist, *d*-tubocurarine, at  $10^{-3}$  mol l $^{-1}$  ( $N = 2$ ) eliminated the suppression of spontaneous



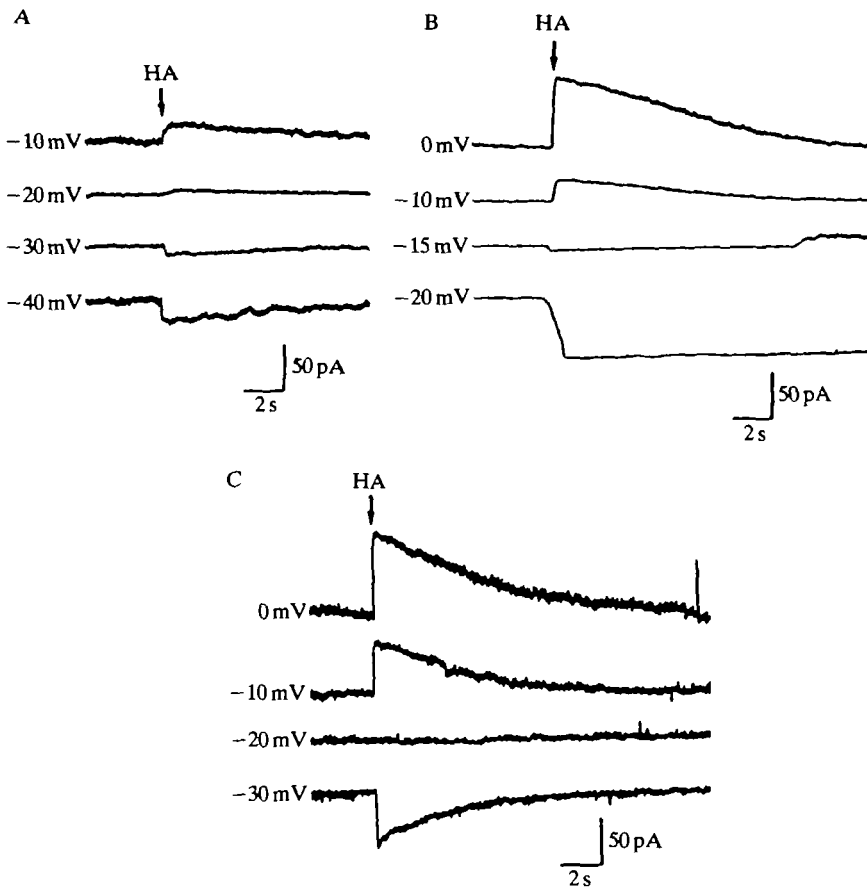


Fig. 7. Whole-cell voltage-clamp recordings of the HA-induced currents from three somata isolated from lobster olfactory receptor cells recorded at different membrane potentials. In each instance,  $10^{-3} \text{ mol l}^{-1}$  HA was ejected onto the soma for either 25 or 100 ms (constant for any one cell). (A) Recording with  $140 \text{ mmol l}^{-1}$  KCl patch solution and normal saline ( $E_{\text{Cl}} = -31 \text{ mV}$ ) resulted in the current reversing polarity at about  $-25 \text{ mV}$ . (B) Recording with  $200 \text{ mmol l}^{-1}$  KCl patch solution and normal saline ( $E_{\text{Cl}} = -24 \text{ mV}$ ) shifted the reversal potential to about  $-12 \text{ mV}$ . (C) Recording in  $140 \text{ mmol l}^{-1}$  patch solution, but using  $\text{K}^+$ -free saline ( $E_{\text{Cl}} = -31 \text{ mV}$ ), failed to shift the reversal potential from that observed in A.

activity induced by  $10^{-5} \text{ mol l}^{-1}$  HA when the antagonist and HA were premixed and presented simultaneously to the preparation (Fig. 8). Increasing the concentration of HA overcame the cimetidine block (Fig. 8,  $N = 1$ ). The vertebrate  $\text{H}_1$  antagonist, pyrilamine, at  $10^{-3} \text{ mol l}^{-1}$  and presented in the same way as cimetidine, was without effect ( $N = 4$ ), as was the vertebrate  $\text{H}_3$  antagonist, burimamide ( $N = 3$ ). Preincubating with the antagonists for 3 min prior to presentation of HA did not affect the results, nor did any of the antagonists have

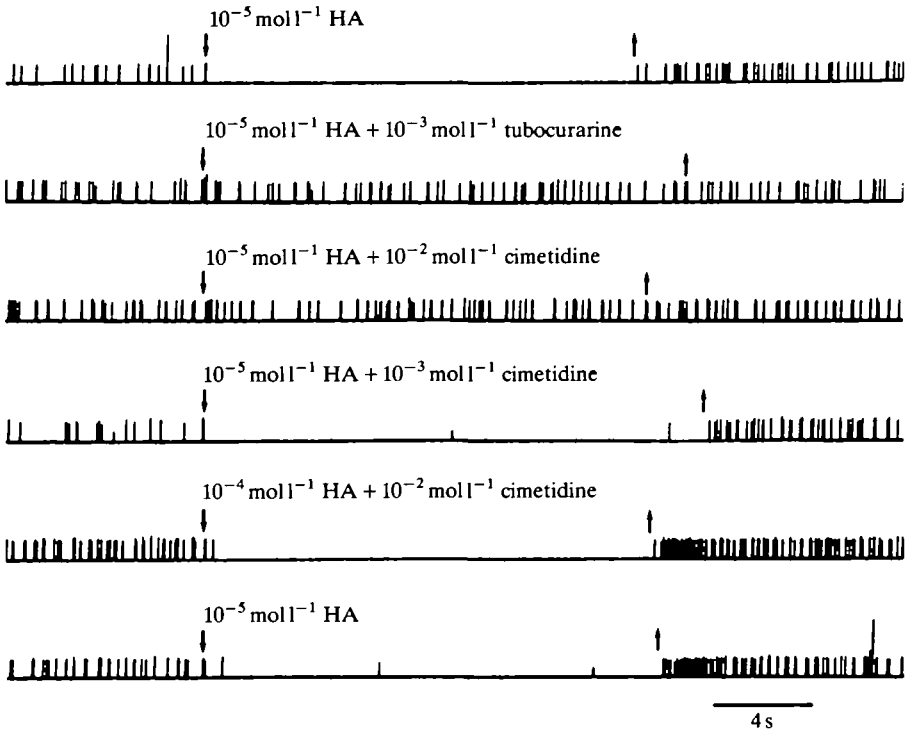


Fig. 8. Extracellular recordings of spontaneous activity from a lobster olfactory receptor cell showing the pharmacology of HA-induced suppression. In all instances drugs were manually pipetted onto the soma at the downward arrow and removed by saline rinse at the upward arrow. Top trace,  $10^{-5} \text{ mol l}^{-1}$  HA suppressed the spontaneous activity of the cell. Second trace,  $10^{-3} \text{ mol l}^{-1}$  *d*-tubocurarine blocked the action of  $10^{-5} \text{ mol l}^{-1}$  HA. Third and fourth traces,  $10^{-2} \text{ mol l}^{-1}$ , but not  $10^{-3} \text{ mol l}^{-1}$ , cimetidine blocked the action of  $10^{-5} \text{ mol l}^{-1}$  HA. Fifth trace, increasing the concentration of HA from  $10^{-5}$  to  $10^{-4} \text{ mol l}^{-1}$  overcame the block otherwise induced by  $10^{-2} \text{ mol l}^{-1}$  cimetidine. Last trace, the initial effect of  $10^{-5} \text{ mol l}^{-1}$  HA persisted.

any effect when applied by themselves. The vertebrate  $\text{H}_2$  agonists impromidine ( $2 \times 10^{-3} \text{ mol l}^{-1}$ ) and dimaprit ( $10^{-4}$  or  $10^{-3} \text{ mol l}^{-1}$ ) failed to mimic the suppressive effect of HA.

HA suppressed the excitability of olfactory receptor cells in the clawed lobster in the same manner as that described for the spiny lobster. Somatically applied HA inhibited spontaneous activity in the same rapid, slowly adapting manner (Fig. 9A vs Fig. 2,  $N = 7$ ). HA also transiently depolarized the cells in the normal recording conditions with an associated increase in membrane conductance (Fig. 9B vs Fig. 4,  $N = 15$ ). Elsewhere, we have shown that HA alters membrane conductance in the clawed lobster as the result of direct gating of a chloride channel (T. S. McClintock & B. W. Ache, in preparation).

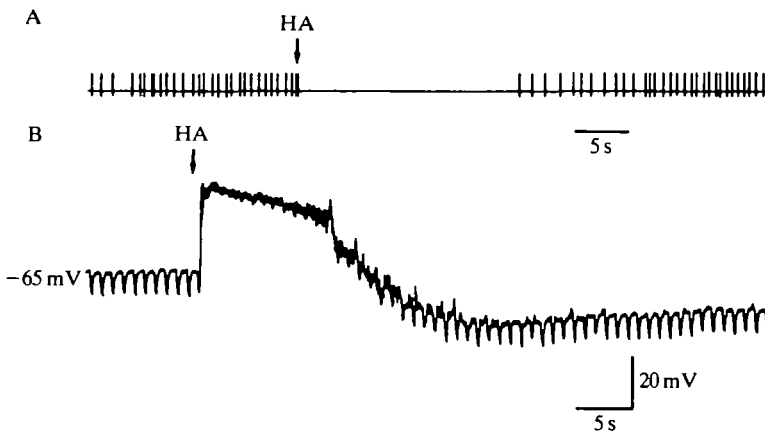


Fig. 9. Recordings from two clawed lobster olfactory receptor cells. (A) Extracellular recording showing that pressure-ejecting  $10^{-4} \text{ mol l}^{-1}$  HA onto the soma (900 ms) suppressed the spontaneous activity of the cell. (B) Whole-cell current-clamp recording showing that pressure-ejecting  $10^{-4} \text{ mol l}^{-1}$  HA onto the soma (600 ms) depolarized the cell with an associated increase in membrane conductance. Recorded with high-KCl ( $200 \text{ mmol l}^{-1}$ ) patch solution and normal saline ( $E_{\text{Cl}} = -24 \text{ mV}$ ).

### Discussion

The presence of a HA-activated chloride conductance on the somata of the olfactory receptor cells of two species of lobsters strongly suggests that the cells are the target of some modulatory process. Action potentials are thought to originate at or near the axon-soma junction in these cells (Schmiedel-Jakob *et al.* 1989). Since the soma possesses steady-state chloride channels (T. S. McClintock & B. W. Ache, in preparation), it is likely that  $E_{\text{Cl}}$  falls close to the resting membrane potential. By increasing chloride conductance, HA would effectively clamp the soma at or near the resting potential and reduce the probability that an invading receptor potential would bring the spike generator to threshold. This mechanism would also reduce the probability of spontaneous discharge and thus is consistent with the extracellular data. It is also consistent with earlier reports that HA activates a chloride conductance to inhibit or hyperpolarize neurones in spiny lobsters (Claiborne & Selverston, 1984) and other invertebrates (*Aplysia*, Weinreich, 1977; Gruol & Weinreich, 1979; McCaman & Weinreich, 1985; fly, Hardie, 1987).

Several of the pharmacological results are consistent with the hypothesis that HA acts *via* a receptor: the effect appears to be specific to HA, cimetidine completely and reversibly blocks the action of HA, and the cimetidine block can be overcome by increased concentrations of HA. A necessary caveat is that the millimolar concentration of the antagonists required for complete blockade allows for possible non-specific effects. A reasonable, alternative explanation, however, is that the effect we report is receptor-mediated, but that the vertebrate-derived HA antagonists presently available have a low affinity for invertebrate HA

receptors, as shown by the high concentrations of cimetidine required to block HA-induced effects in flies (Hardie, 1988) and barnacles (Stuart & Callaway, 1988). If so, then the pharmacological profile of the putative HA receptor in lobster olfactory cells is not consistent with that of classical H<sub>1</sub>, H<sub>2</sub> or H<sub>3</sub> types of vertebrate HA receptors (Schwartz *et al.* 1986), or with that of other invertebrate HA receptors mediating chloride conductances that are blocked by tubocurarine, but not by cimetidine (*Aplysia*, Gruol & Weinreich, 1979; spiny lobster, Claiborne & Selverston, 1984). The HA receptor in lobster olfactory receptor cells may belong to an emerging class of HA receptors with a combined cholinergic and histaminergic pharmacological profile (Hardie, 1988) that appears to be associated with a chloride conductance (Hardie, 1987; Zettler & Straka, 1987). Cimetidine block has also been associated with HA-induced increases in potassium permeability in *Aplysia* neurones (Carpenter & Gaubatz, 1975; McCaman & Weinreich, 1982, 1985).

Our findings provide some insight into the possible functional significance of the HA-induced suppression of excitability. The lack of any coupling between dendritic and somatic sensitivity to HA suggests that the somatic receptors are not involved in 'intravascular' chemoreception (Maruniak *et al.* 1983). That almost all cells tested were suppressed is indicative of a modulatory role for HA, but the relatively high threshold, rapid onset and rapid decay of HA's action in our system does not support a blood-borne effect (Kravitz, 1988). These actions of HA are more suggestive of a synaptically mediated effect, an alternative supported by our finding (T. S. McClintock & B. W. Ache, in preparation) that HA directly gates a chloride channel in these cells. As yet, however, there is no evidence for peripheral synapses in the lobster olfactory pathway (Spencer & Linberg, 1986; Grünert & Ache, 1988). It is still possible that synaptically mediated modulation could occur at the central termini of these primary receptors. Both immunocytochemical and biochemical evidence localize HA to the brain region in which the receptor cells terminate (E. A. Orona, B. A. Battelle & B. W. Ache, unpublished data). In this context, HA receptors on the soma could possibly represent stray postsynaptic receptors. The alternative that somatic HA receptors are autoreceptors (Arrang *et al.* 1985) is minimized by our inability to obtain biochemical or immunocytochemical evidence that the receptor cells are histaminergic (E. A. Orona, B. A. Battelle & B. W. Ache, unpublished data).

Further experimentation is necessary to resolve the functional significance of somatic sensitivity to HA in lobster olfactory receptor cells. Nevertheless, the possibility that the cells may be subject to modulation opens up a new level of understanding in the organizational complexity of the olfactory pathway in lobsters and possibly other species.

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