HEAD WAVING IN APLYSIA CALIFORNICA

I. BEHAVIOURAL CHARACTERIZATION OF SEARCHING MOVEMENTS

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Summary

Searching is an important component of several types of goal-directed behaviour. In soft-bodied animals, searching behaviour can appear quite complex because the range of body movement is not constrained by joints, limbs or muscles with discrete areas of origin and insertion. In addition, animals exhibiting this type of behaviour utilize their maximum freedom of movement. In this paper, we describe the head-waving searching behaviour of the sea hare Aplysia californica by characterizing patterns of movements and the changes in body shape that underlie these movements. A bout of head waving consists of a series of discrete movements separated by pauses. Each discrete movement lasts 4–10 s and is directed either horizontally or vertically with respect to the stationary part of the animal. Large movements, such as bending from the extreme right to the extreme left, consist of a series of these shorter movements separated by pauses lasting 1–2 s. In all head-waving movements, the transverse axis of the head is kept relatively parallel with the substratum. Thus, vertical movements require only bending of the body, whereas horizontal movements require twisting of the body, particularly when the posture is more erect. During head waving, the anterior two-thirds of the body is free to move, but most of the bending occurs immediately posterior to the head region. There is no periodicity of movements within a bout of head waving, although isolated instances of repeated movements are sometimes observed. Therefore, although the individual movements during head waving are relatively simple, the absence of a patterned sequence accounts for the complexity of the overall behaviour. These observations both constrain models of the neural organization of head waving and provide criteria for categorizing head-waving movements in further behavioural and physiological studies.

Introduction

A fundamental goal of neuroethology is to understand the principles of neural circuit structure and function responsible for organizing and controlling different types of animal behaviour. Behaviour changes through development and learning, so knowledge of these

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neural circuits is necessary for the analysis of cellular mechanisms that underlie the changes. In the last two decades, considerable attention has been focused on highly stereotyped behaviour patterns, such as graded reflexes, fixed action patterns and rhythmic movements (Camhi, 1984; Carew and Kelley, 1989). However, many types of behaviour are not easily described as a specific single pattern of movement because animals may use different movements to produce the same functional result, or the specific form of a movement may depend on the animal’s initial posture, orientation and sources of external stimuli. These more complex behaviours include appetitive, searching behaviours in which an animal moves its head and/or sensory appendages to receive and orient to specific stimuli in the environment. Besides being important components of an animal’s behavioural repertoire, these behaviours may also show forms of plasticity that are difficult to demonstrate or produce in simpler, more stereotyped behaviours.

The gastropod mollusc Aplysia californica has served as a useful model system for developing our understanding of the organization and plasticity of several simple withdrawal, defensive and locomotory behaviours at the cellular and molecular levels (Kandel, 1979). In addition to these relatively stereotyped behaviours, Aplysia exhibits more complex behaviours that are also amenable to cellular analysis. One such behaviour is head waving, which plays an important role in the appetitive phase of several complex motor acts, such as feeding (Kupfermann, 1974), egg laying (Cobbs and Pinsker, 1982), mating (Leonard and Lukowiak, 1986) and locomotion (Kupfermann and Carew, 1974).

An analysis of head waving is also an important step in defining the cellular and molecular bases of operant conditioning. Operant conditioning is a form of learning in which an animal learns to associate a particular response with the consequences of that response; either a reward or punishment. In Aplysia, if an aversive stimulus such as bright light is delivered while the animal is head waving to one side of its body during a ‘training’ period, the animal will spend more time bending to the opposite side during a subsequent ‘testing’ period (Cook and Carew, 1986). Thus, the animal learns to associate bending its body in a certain direction with a negative outcome of that response: the occurrence of an aversive stimulus. To identify the cellular and network changes that are responsible for the operant conditioning, it is important to know more about the central connections of sensory neurones mediating the reinforcement as well as the interneuronal network responsible for commanding, coordinating and controlling the head-waving behaviour. Cook and colleagues have begun an analysis of the reinforcement pathway (Cook and Carew, 1989c; Cook et al. 1991), but nothing is known about the central neural control of the head-waving response that is modified.

The present series of papers was therefore directed at a systematic analysis of a complex behaviour in Aplysia, head waving, for two interrelated reasons. First, understanding the neural architecture responsible for this behaviour could contribute to the neuroethological goal of gaining insights into mechanisms underlying complex adaptive behavioural responses. Second, such an understanding could elucidate important cellular loci and mechanisms that may be involved in a fundamental form of associative learning, operant conditioning. In this and the following two papers (Kuenzi and Carew, 1994a,b) we will establish a basis for the analysis of head waving at a cellular level. In the present paper, we describe the behaviour, including the topography of head-waving
movements and patterns of movement during a bout of head waving. In the second paper (Kuenzi and Carew, 1994a), we describe the musculature of the body wall and the spatial and temporal patterns of activation of these muscles during head waving. In the third paper (Kuenzi and Carew, 1994b), we use the anatomy of the central nervous system to begin locating interganglionic interneurones that contribute to the execution and coordination of head waving.

To describe head waving in Aplysia we will consider two questions. First, from a behavioural perspective, are there any patterns in the timing and direction of movements that could provide a basis for identifying basic components of the overall behaviour? Second, from a kinematic perspective, the body plan of Aplysia (i.e. soft-bodied and fluid-filled) endows it with almost unlimited freedom of movement, including extension along its long axis, bending in all directions perpendicular to this axis and torsion about the long axis. How does the animal utilize this freedom during head waving?

**Materials and methods**

**Animal maintenance**

*Aplysia californica* were obtained either from the wild, through a commercial supplier (Marinus, Inc., Long Beach, California, USA), or from a laboratory culture maintained at the University of Miami, USA. All animals were held for at least 48 h in a 530 l capacity fibreglass aquarium filled with recirculating artificial sea water cooled to 16 °C, and fed with the seaweed *Gracillaria* sp.

**Behavioural procedures**

To identify patterns of movement, animals (40–60 g) were observed and videotaped while head waving in an acrylic aquarium (25 cm×25 cm×25 cm) filled with water from the animal’s home tank (Fig. 1). The water was not aerated during the observation sessions in order to prevent water currents from interacting with the animal’s own muscle activity in producing movement. However, the aquarium water was changed between sessions (which lasted no longer than 30 min).

Two behavioural preparations were utilized. (1) In the suspended animal preparation (Cook and Carew, 1986), an animal was suspended in the seawater observation aquarium by its parapodia. A stainless-steel hook attached to a thread was inserted into each parapodium just anterior to its midpoint. The free end of the thread was then attached to a crossbar above the aquarium. This isolation from a solid substratum reliably induces head waving and has proved useful in previous studies of operant conditioning of the head-waving response (Cook and Carew, 1986, 1989a–c) and phototactic modulation of head waving (Kuenzi and Carew, 1991). (2) In the substratum-attached preparation, an animal was allowed to crawl onto a circular platform (diameter smaller than the animal’s fully contracted body length), which was separated from the walls of the aquarium by a distance greater than one body length (Fig. 1). In this situation, an animal typically begins head waving soon after extending its head over the edge of the platform, using the posterior part of its foot as a holdfast. This preparation provides a more stable frame of reference for kinematic analysis. To keep the animal from touching the walls or floor of
the aquarium, the platform was held onto one end of a tubular pedestal by suction applied to the other end. Once the animal had established a holdfast on the platform, the platform and animal could be rotated together in order to film either a front or a side view. The dorsal view of the animal was provided by an angled mirror above the aquarium. The corresponding vertical angle ($\theta_v$) was measured from either the side view, as shown, or the front view.

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**Behavioural quantification**

Head-waving movements were classified into seven components (see Results), and each of the components was assigned to a key on a computer keyboard. The sessions of videotaped behaviour were scored by an observer entering the appropriate keystroke at the end of each movement. The list of keystrokes from each session was saved on a
computer disk for later analysis. Summaries of measured values, such as the direction of movement (see Fig. 5), are presented as the between-animal mean ± standard error of the mean and are based on repeated within-animal measures.

Measurement of body movement

The angle between the holdfast and the body column/head was measured in both the horizontal plane ($\theta_H$) and the vertical plane ($\theta_V$; Fig. 1). A one-turn, linear potentiometer in a voltage divider circuit was used to measure each of these angles in real time from the video playback. The potentiometer was mounted on a hand-held acrylic plate, and a line was etched from the centre of the potentiometer to the edge of the plate. This line was positioned parallel to the longitudinal axis of the animal’s holdfast. A 6 cm long acrylic rod with one end attached to the shaft of the potentiometer was used as a pointer with which an observer manually tracked the orientation of the anterior part of the body relative to the holdfast (Fig. 1). The output voltage was fed into a microcomputer through an analogue-to-digital converter, and a data acquisition program sampled the signal at either 1 or 2 Hz. The mid position of the potentiometer corresponded to the 0˚ position, and deviations from this (higher or lower resistance) corresponded to movement of the body column/head in either the horizontal or vertical direction. An electromyogram was recorded from the body wall muscles as described in Kuenzi and Carew (1994a). The angles $\theta_H$ and $\theta_V$ were measured separately, and the resulting traces were aligned by comparing the electromyographic record, which was synchronized with the video tape and replayed with each measurement. Although this method was appropriate for the qualitative purposes of this study, it was not sufficient for acquiring precise, quantitative measures. However, it was more efficient than frame-by-frame digitization of the video image and more accurate than digitization because the experimenter was able to compensate for animals that had an unsteady attachment to the substratum and for movements of animals that were not aligned perfectly with the camera.

Results

Overview of head waving

An animal may begin the undirected, searching type of head waving (the term ‘head waving’ has also been applied to two other types of movements, see Discussion) spontaneously, i.e. with no observable stimulus, or after contact with a sprig of seaweed, another animal or, in some cases, a glass rod. The form of the behaviour does not depend on the type of initial stimulus, as long as there is no further contact with solid objects. After a local withdrawal of the site of stimulation, the animal releases the anterior end of its foot from the substratum, lifts its head and moves through the water by bending and twisting the anterior part of its body. Early in a bout, a large portion of the foot is attached to the substratum (Fig. 2A), but during the course of the bout the animal extends its reach both by releasing successively posterior regions of the foot from attachment and by lengthening (extending) the unattached body (Fig. 2B,C). Eventually, only the tail region remains attached; thus, the head-waving motion allows the animal to sweep its head through any point within approximately one body length of the holdfast. If no objects are
encountered in this area, the bending movements will bring the propodium down to contact the substratum adjacent to the holdfast, and the animal will make attachment as during locomotion. Several cycles of locomotion follow before the animal either becomes quiescent or begins another bout of head waving at a new site.

A second method of eliciting head waving is to isolate the animal from contact with the substratum by suspending it in water. However, because the substratum-attached animal provides a more stable frame of reference, this preparation was used for the kinematic analysis. We then compared these movements with the head-waving movements of suspended animals for congruity with earlier studies of operant conditioning (Cook and Carew, 1986) and biomechanics (Kuenzi and Carew, 1988, 1994a).

Frame of reference

Although head waving is a three-dimensional movement, the change in length of the animal can be described as a slow monotonic extension, as described above. The correspondence between elongation and the other movements was not analyzed directly, but the difference in time course between elongation and the slowest vertical or horizontal movements suggests that they are independent and that head waving can be approximated as a two-dimensional behaviour.

Fig. 2. Extension during head waving. Tracings of video images illustrate the progression of extension that was observed early in a bout (A) and at later stages of head waving (B,C). Examples were selected from tapes of different animals. In each panel, the top view of the animal is above and a side view is below. The division of the body into different regions is also shown: H, head; BC, body column; Hld, holdfast. The ‘substratum’ platform is represented by a partial circle in the top view and a rectangular shape in the side view. Heavy shading (top view) indicates the area of the foot attached to the substratum. at, anterior tentacles; ppd, parapodia; rh, rhinophores.
For the purpose of description, the orientation of the holdfast was used to define the ‘resting’ orientation of the more anterior, mobile part of the body (Fig. 1). Thus, when the longitudinal axis of the anterior part of the body is in line with that of the holdfast, the horizontal $\theta_H$ and vertical $\theta_V$ body angles will both be $0^\circ$. Positive vertical angles correspond to dorsal flexion, and positive horizontal angles correspond to bending to the right.

**Types of movement**

A detailed video analysis of head-waving bouts from four animals, and further behavioural observations of videotaped sessions from over 30 other animals, revealed three common features, which will be discussed below: (1) head waving involves discrete movements, which are primarily directed horizontally or vertically; (2) the topography of head-waving movements involves both bending and twisting of the body; and (3) most of the movement is in the anterior region, or the head.

**Discrete movements**

A bout of head waving consists of a series of discrete movements that are defined as a brief acceleration of the body, or a burst of movement, followed by deceleration or a pause. Successive bursts of movement were either in the same direction, making what appeared to be a larger movement, or in different directions. In most cases, the direction of motion was biased towards either horizontal or vertical movement. These features are illustrated in a continuous, 75 s segment of a head-waving bout presented in Fig. 3A,B. The lines between points indicate the magnitude (length of the line) and direction (orientation of the line) of the average velocity of the anterior part of the body between the two time points. In this figure, horizontal movements are indicated by open arrows, vertical movements by filled arrows and diagonal movements by double arrowheads. Bursts of movement occurred at intervals of 4–10 s. In Fig. 3, these appear as widely spaced points, whereas the intervening pauses appear as clusters of points. Pauses occur both at the transitions between horizontal and vertical movements and at intervals during the longer movements in one direction. The occurrence of movement bursts within longer movements is better illustrated in Fig. 4, which shows the horizontal component of anterior body orientation as a time series in the upper trace and the corresponding average velocity in the lower trace. The vertical angle ($\theta_V$) was near $0^\circ$ for the duration of this record. Although the two bends from $0^\circ$ to the left and back appear to be roughly sinusoidal, the corresponding slow component of the velocity was small compared with the spikes that indicate short rapid movements.

The data in Fig. 3 also suggest that, despite the nearly unlimited freedom of movement available to the animal, its movements are strongly biased towards relatively few directions. To show this more clearly, the directional component of the body angular velocity was computed as the average velocity during 1 s sampling intervals, and these values were sorted into $10^\circ$ bins and combined across animals. This distribution (Fig. 5) was markedly skewed towards vertical movements (dorsal and ventral) and horizontal movements (right and left), with relatively little motion in the diagonal directions. The data in Fig. 5 do not discriminate between active (acceleration phase) and passive
(deceleration phase) movements. From Fig. 3 and other behavioural observations, it was clear that the active movements were even more strongly biased towards the horizontal and vertical directions. Taken together, these observations suggest that the complex

Fig. 3. An example of head-waving movements. The horizontal and vertical components of body orientation ($\theta_H, \theta_V$) are plotted on their respective axes to simulate a frontal view of the path of an animal’s head during head waving (note, however, that the axes are scaled in degrees rather than distance). The sampling interval was 1 s, and the numbers at the beginning and end of each trace indicate the time, in seconds, from the beginning of the record. The striped bar at the origin indicates the position of the platform. A and B form a continuous record, with the last six points in A being repeated as a dashed line in B for reference. Arrows indicate the direction of putative discrete movements as discussed in the text: open, horizontal; filled, vertical; double arrowhead, diagonal.
Head-waving movements of Aplysia

movements of head waving can be described in terms of discrete movements in relatively few directions.

Topography

The above analysis describes the motion of a terminal point on the head with respect to a fixed point in the holdfast, but does not take the underlying changes in the body shape into account. Although these movements could be produced by bending the longitudinal axis, closer examination of head-waving animals and videotape records showed that the topography of head waving was more complex. Specifically, the pitch of the head (orientation of the transverse axis of the head with respect to the horizontal plane of the holdfast) remained constant or rotated against the direction of movement. For example, while bending to the left, the animal’s head rotated slightly so that the left side of the propodium was elevated with respect to the right side (Fig. 6A). When the anterior of the animal’s body was elevated during the horizontal motion, this ‘stabilization of the head’ was opposite to the change in head orientation expected if the animal were simply to bend its body (Fig. 6B). Animals did make head-waving movements similar to that shown in Fig. 6B (described as the arch movement below), but this was relatively rare.

Fig. 4. Head-waving movements are brief. Bursts of movement are illustrated in this trace of lateral sweeps to the left and centre. Pauses appear as shoulders in the head angle trace (A) or as near-zero values in the corresponding velocity trace (B). The horizontal body orientation ($\theta_{ho}$, Fig. 1) was digitized at 2 Hz and the magnitude of the velocity was computed using the formula: velocity = ($\theta_{hi,t} - \theta_{hi,t-1}$)/0.5, where $t$ is an index of time. The arrows indicate some periods of more rapid movement.
To facilitate discussion of actual head-waving movements, we have developed a catalogue of movements by combining the direction of movement and the change of body shape involved. A head-waving bout can be described as a sequence of discrete movements, such as those indicated by arrows in Fig. 3, rather than as pairs of horizontal and vertical body angles. Examples of the different categories, or component movements, are illustrated by the time-lapsed overlays of video images shown in Fig. 7.

Vertical movements involve either dorsal or ventral flexion of the body, but without twisting, and these are described as lifting and depression of the body (Fig. 7E,F respectively). In addition, a third category, tucking (not illustrated), includes a behaviour in which the anteriormost part of the body is sharply flexed downwards. This movement brings the propodium towards the anterior edge of the holdfast for searching the area near the holdfast.

The topography of horizontal movements depends on the vertical angle of the body during the movement. When the vertical body angle was low, as in a lateral sweep, bending was the most significant component, although some twisting of the head was observed (Fig. 7A). When the head was moderately elevated, as in a helical sweep

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Fig. 5. Head-waving movements are primarily in the dorsoventral and horizontal directions. The average directional component of body angular velocity was calculated from data such as those presented in Fig. 3 (1 s sample interval) using the formula: angular velocity = $\tan^{-1}[(\theta_{t1} - \theta_{t1-1})/((\theta_{t1} - \theta_{t1-1})]^2$. The directions of movement were grouped into 10° bins and converted to percentages of the given bout. The dashed circle represents a value of approximately 6%. Between-animal means and standard errors (four animals) were computed as described in the Materials and methods section.
or fully erect, as in an upright pivot (Fig. 7C), more torsion was used to stabilize the pitch of the head. These three divisions correspond to vertical angles of up to 30˚ for the lateral sweep, 30–70˚ for the helical sweep and over 70˚ for the upright pivot (see, for example, the open arrows in Fig. 3). Simple lateral bending of the body while the animal was in an elevated posture turns the head towards the substratum so that the body forms an arch. This was occasionally observed and was named arching (Fig. 6B).

Diagonal movements show the same head stabilization as that seen in other types of movement, but are otherwise very diverse in form, resembling either vertical or horizontal movements to different extents. For simplicity, they have been classed together as diagonal sweeps (Fig. 7D).

The overall incidence of the different movements was estimated from the behaviour of four animals, with three or four sessions being analysed for each animal and each session, including 30–100 transitions from one component to another. The contribution of each movement to a bout was computed within each bout, and these were averaged across bouts for each animal. Finally, the animal scores were combined to give an overall mean and standard error (N=4). Horizontal movements were relatively evenly divided between lateral sweeps (11±2.4%), helical sweeps (7±2.3%) and upright pivots (7±2.3%), which together accounted for 25% of all movements. Vertical movements were also evenly divided between lifts (18±3.7%) and depressions (22±1.2%), which together accounted for 40% of the movements. The diagonal movements alone accounted for 21±3.2%, whereas the tuck and arch movements were rare, together contributing only 3±1.3%. The remaining 11% included periods when the animal was motionless (which could occur while the animal was in any posture) and when it transiently contacted the walls of the aquarium.

Movement of the head

The final component of head-waving topography was the position of bending along the
Fig. 7. Component head-waving movements. Each component movement described in the text is illustrated by a series of three or four tracings from a video monitor. The interval between each trace is approximately 3 s. The top image of each panel is the dorsal view of the animal, and the lower image is the direct frontal view (A–D) or lateral view (E,F). Thus, the left side of the panel is the animal’s right for both images. For each component movement, the final position is shown as an entire thick-lined outline. The essential features of earlier traces are shown with fine lines, which are dashed (second trace) or dotted (earliest trace) where the image overlaps the final trace. The arrows indicate the direction of movement.
longitudinal axis of the animal. Although the body of *Aplysia* is not segmented anatomically, we often observed a point of inflection, or local bending, between the parapodia and the rhinophores (Fig. 8, open arrowheads). The body of an animal may therefore be divided into a head region and a body column region that extends from the inflection to the holdfast (Fig. 2). The movements shown in Fig. 8 illustrate two features of independent head and body column movements. First, head-waving movements generally began with bending of the head in the direction of motion. In Fig. 8A, the animal was reversing its direction of motion from a right lateral position to begin a leftward bend back towards the centre. This movement began with straightening of the body, followed by bending of the head in the opposite direction while the body column angle was still bent to the right. Second, head movements were more rapid and more frequent than body column movements; for example, the head could reverse direction one or more times during a single general body movement (Fig. 8B).
Movement patterns

Although the traces of body orientation angles, such as that shown in Fig. 4, suggest that the behaviour may have an underlying cyclic structure, longer movements were generally not repeated. When consecutive, similar movements were observed, the number of cycles was low (2–3); however, it was interesting that their period was consistently of the order of 30 s for both horizontal and vertical movements. This agrees well with the more continuous side-to-side bending observed in suspended animals (Kuenzi and Carew, 1991). To test for the presence of a cyclic pattern in head waving, we computed the autocorrelation function for 12 bouts of the behaviour. In all cases, however, the function decayed smoothly to zero without showing any periodicity.

Head waving of suspended animals

Adult *Aplysia californica* are almost always attached to the substratum by some portion of their foot (Kupfermann and Carew, 1974; Leonard and Lukowiak, 1986). Although juvenile animals typically respond to detachment from the substratum by opening their parapodia (which slows their descent), adults close their parapodia and flatten their body laterally when detached (Kupfermann and Carew, 1974). Prolonged suspension in still water, however, consistently elicits movements similar to those of head waving (Cook and Carew, 1986). Analogues of all the component movements illustrated in Fig. 7 and described above can be identified, including the different types of horizontal movements. The transverse axis of the head was stabilized with respect to the midsection of the body.
(near the suspension lines), and the body could be divided into head, body column and tail on the basis of relative movement (Fig. 9). Furthermore, an animal’s behaviour was not noticeably altered when its tail chanced to attach to the rear of the aquarium, where it then served as a holdfast.

Discussion

Three types of head waving can be distinguished on the basis of duration and orientation to external stimuli. First, in the ‘undirected head waving’ that is the subject of our study (‘phase 1’ of appetitive feeding behaviour; Teyke et al. 1990), the posterior portion of the ventral surface of the foot grips the substratum to form a holdfast, while the anterior body is held erect and slowly bent and twisted so as to sweep the head repeatedly through the water around the animal (Kupfermann, 1974; Kupfermann and Carew, 1974; Leonard and Lukowiak, 1986; see below). Typically, this begins after a brief arousing stimulus and, when the stimulus is removed, may continue for tens of minutes with no bias in the direction of bending over the course of the bout (Cook and Carew, 1986). Second, ‘directed head waving’ (including ‘phase 2’ of feeding; Teyke et al. 1990) is a transient response to contact with an object, such as a glass rod or piece of seaweed, in which the body is bent rapidly towards the stimulus. This typically occurs during a bout of undirected head waving and serves to centre the animal’s mouth and propodium on the object. Third, in the laboratory, animals can be made to exhibit a more prolonged ‘taxis’ response to certain external stimuli: that is, the back-and-forth head movements continue, but with a bias towards or away from the direction of stimulation. This has been shown for the response to light in some situations (Kuenzi and Carew, 1991), and under natural conditions it may play a role in foraging (Frings and Frings, 1965). Together, these head-waving behaviours allow an animal to locate and explore objects in its environment.

Undirected head waving is a more complex behaviour than those that have served as simple systems in neuroethology. Contributing to this complexity is the fact that, both in the laboratory and in the field (Kupfermann and Carew, 1974), the animal moves in three dimensions, with few constraints on its use of space, as might be imposed by a rigid, articulated skeleton for example. In addition, there are no repeated patterns of body movement on which to establish the phase of muscle or neural activity. Therefore, to help in the cellular analyses of this complex behaviour, the kinematic and behavioural analysis presented in this paper identified spatial and temporal patterns of movement that will form a basis for further neurophysiological investigation of both head waving itself and the operant conditioning of head waving.

We have found that head waving can be described relatively simply and may be governed by relatively simple rules. First, elongation of the animal is slow, compared with the bending and twisting movements, and can be described as a monotonic extension of the body throughout a bout of head waving. Therefore, on the time scale of the more rapid movements, the behaviour is essentially two-dimensional. Second, the duration of a movement is 4–10 s, even though changes in direction of movement may occur at longer intervals. Longer movements in one direction consist of a series of brief, discrete movements that are of the same duration as the shortest movements. Third, movements
are biased towards the horizontal and vertical directions, with comparatively little diagonal movement. Finally, there are at most two sites of local bending: (i) at the junction of the head and body column; and (ii) at the junction of the holdfast and body column. These observations both constrain models of the neural organization of undirected head waving and provide criteria for categorizing head-waving movements in further behavioural studies.

The rarity of head-waving movements diagonal to the horizontal and vertical axes is surprising, considering the fact that there are no corresponding physical restraints on the animal’s freedom of bending movement (there are longitudinal muscles all around the circumference of the animal’s body, allowing the animal to bend in any direction; Kuenzi and Carew, 1994a). In a homogeneous environment, an animal with a hydrostatic skeleton should make approximately equal use of its entire range of movement. In undirected head waving, however, animals tended to switch abruptly between horizontal and vertical movements (Fig. 3), with few movements that could be classified as diagonal, and spent relatively little time moving in a diagonal direction (Fig. 5). A general model for head waving could be based on discrete movements generated by a few central programmes specifying different directions of movement. These would coordinate activation of muscles and integrate feedback from gravity receptors to produce the correct topography. Although it is premature to make specific hypotheses about the neural coordination of head waving, the distinctness of horizontal and vertical movements suggests that they may be controlled by separate pattern-generating circuits. This model, albeit quite general, is attractive for the cellular analysis of operant conditioning of head waving, as the initial studies have focused on the horizontal component of the behaviour as the operant response (Cook and Carew, 1986). The present observations support the assumption that it may be possible to isolate the horizontal component for future physiological studies.

Opisthobranch molluscs have provided useful model systems for exploring the cellular basis of behaviour. However, the undirected head waving described in the present paper does not fit neatly into the traditional categories of behaviour for such analyses. It is neither strongly rhythmic, such as locomotion or feeding (Getting, 1983; McPherson and Blankenship, 1991; Rosen et al. 1991), nor directed towards a specific stimulus, as is directed head waving (Teyke et al. 1990). It more closely resembles the complex, adaptive responses exhibited by many species of animals during searching behaviours such as foraging and local terrestrial exploration. This paper shows that the complex movements of a searching behaviour, undirected head waving, can be distilled into a few types of movement, each with a relatively simple organization. In addition, it provides a framework for further analyses of this behaviour using other behavioural techniques. The following papers (Kuenzi and Carew, 1994a,b) describe the body-wall muscles that produce these movements, and show how lesioning the interganglionic neural pathways affects the coordination of muscles during head waving.

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