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Vestibular compensation in lampreys: restoration of symmetry in reticulospinal commands

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Summary

Removal of a vestibular organ (unilateral labyrinthectomy, UL) in the lamprey results in a loss of equilibrium, so that the animal rolls (rotates around its longitudinal axis) when swimming. Owing to vestibular compensation, UL animals gradually restore postural equilibrium and, in a few weeks, swim without rolling. Important elements of the postural network in the lamprey are the reticulospinal (RS) neurons, which are driven by vestibular input and transmit commands for postural corrections to the spinal cord. As shown previously, a loss of equilibrium after UL is associated with disappearance of vestibular responses in the contralateral group of RS neurons. Are these responses restored in animals after compensation? To answer this question, we recorded vestibular responses in RS neurons (elicited by rotation of the compensated animal in the roll plane) by means of chronically implanted electrodes. We found that the responses re-appeared in the compensated animals. This result supports the hypothesis that the loss of equilibrium after UL was caused by asymmetry in supraspinal motor commands, and the recovery of postural control in compensated animals was due to a restoration of symmetry.

Key words: postural control, locomotion, vestibular compensation, reticulospinal system, lamprey, *Lampetra fluviatilis.*

Introduction

In all classes of vertebrates, ablation of one vestibular organ (unilateral labyrinthectomy, UL) evokes severe motor disorders that include abnormal position of eyes, spontaneous ocular nystagmus, asymmetry in the head and trunk posture, etc. Over time, these disorders gradually diminish. This process of the recovery of motor functions is usually referred to as 'vestibular compensation' and is considered to be one of the most striking examples of CNS plasticity (for reviews, see Schaefer and Meyer, 1974; Smith and Curthoys, 1989; Curthoys and Halmagyi, 1999; Dieringer, 1995; Vidal et al., 1998). Despite extensive studies of vestibular compensation, neuronal mechanisms causing the different UL-evoked symptoms and of the recovery of motor functions are still poorly understood. The main reason is that the corresponding neuronal networks are extremely complex.

We use a simple biological model – the lamprey, a lower vertebrate (cyclostome), for studying the effect of UL on postural stability, as well as the process of recovery of postural function. The basic design of the lamprey CNS, and especially of the brain stem and spinal cord, is similar to that of higher vertebrates (Nieuwenhuys et al., 1998), but the lamprey presents many more opportunities for analytical studies of the nervous mechanisms for postural control, including studies at the network and cellular levels (Orlovsky, 1991; Macpherson et al., 1997).

A swimming lamprey actively stabilizes the dorsal-side-up orientation of its body by the activity of the postural control system driven by vestibular input (de Burlet and Versteegh, 1930; Ullén et al., 1995a; Deliagina, 1995, 1997a,b). Visual input plays only a modulatory role: a unilateral eye illumination evokes a roll tilt towards the source of light –the 'dorsal light response' (Ullén et al., 1993, 1995b), first described in bony fishes by von Holst (1935).

Because the postural control system in the lamprey is driven primarily by vestibular input, the effect of UL in this animal is most dramatic. After UL, lampreys with intact eyes completely lose equilibrium and during swimming continuously roll toward the damaged labyrinth (de Burlet and Versteegh, 1930; Deliagina, 1995, 1997a). During this period, however, the equilibrium can be temporarily restored by creating an asymmetry in visual input, that is, by illuminating the eye contralateral to UL, or by electrically stimulating the corresponding optic nerve. During the process of vestibular compensation, which lasts a few weeks, the UL animals gradually recover their capacity to maintain equilibrium (Deliagina, 1995, 1997a).

The postural network in the lamprey has been characterized in considerable detail. Important elements of this network are the reticulospinal (RS) neurons (Nieuwenhuys, 1972), which transmit commands for postural corrections from the brainstem to the spinal cord. The RS neurons receive vestibular input through interneurons of the vestibular nuclei (Koyama et al., 1989; Northcutt, 1979; Rovainen, 1979; Rubinson, 1974; Stefanelli and Caravita, 1970; Tretjakoff, 1909). They also receive inputs from other sensory systems as well as from the forebrain, brainstem centers and spinal cord (Deliagina et al., 1993; Viana Di Prisco et al., 1995; Dubuc et al., 1993; Rovainen, 1967, 1979; Wickelgren, 1977). In the spinal cord, the RS neurons affect motoneurons and different classes of interneurons (Brodin et al., 1988; Ohta and Grillner, 1989; Rovainen, 1967, 1974, 1979; Wannier et al., 1995; Zelenin et al., 2001, 2003).

Responses of larger RS neurons to natural vestibular stimulation (roll tilts) and eye illumination were initially investigated *in vitro* (Deliagina et al., 1992a; Orlovsky et al., 1992; Deliagina et al., 1993; Ullén et al., 1996), and recently *in vivo* (Deliagina and Fagerstedt, 2000). These experiments have shown that the majority of RS neurons were activated by contralateral roll tilt; this activation was mainly due to excitatory input from specific groups of contralateral vestibular afferents (Deliagina et al., 1992b). A unilateral visual input evoked excitation of the ipsilateral RS neurons and inhibition of the contralateral ones.

Subsequent studies (Deliagina and Pavlova, 2002) have shown that UL caused a dramatic asymmetry in the responses of RS neurons to roll tilts: the responses persisted in the ipsilateral RS neurons and disappeared in the contralateral ones. It was also found that illumination of the eye contralateral to the UL resulted in a restoration of symmetry in the bilateral activity of the RS system. Since illumination of this eye also leads to a restoration of equilibrium in non-compensated UL lampreys (Deliagina, 1997b), it was suggested that the loss of equilibrium and continuous rolling in UL lampreys is caused by the asymmetry in descending RS commands, and a recovery of postural control occurs as a result of restoration of symmetry in these commands (Deliagina, 1997b; Deliagina and Pavlova, 2002).

The goal of the present study was to test this hypothesis. For this purpose, by means of chronically implanted electrodes, we recorded responses to roll tilts in the left and right RS neurons in fully compensated animals. These data were compared to the control data, that is, to the vestibular responses recorded in non-compensated animals soon after UL (Deliagina and Pavlova, 2002). These experiments have shown that the ability of lampreys to maintain equilibrium is associated with the presence of vestibular responses in RS neurons on the side contralateral to the UL. These results support the hypothesis that a recovery of postural control impaired by UL is the result of restoration of symmetry in the supraspinal motor commands.

A brief account of this study has been published as an abstract (Pavlova and Deliagina, 2002b).

Materials and methods

Experiments were carried out on six adult (25–35 cm in length) lampreys (*Lampetra fluviatilis* L.), which were kept in an aerated freshwater aquarium at 7°C, with a 12 h:12 h L:D cycle. All experiments were approved by the local ethics committee (Norra Djurförsöksetiska Nämnden).

Electrodes

The activity of RS neurons was recorded from their axons in the spinal cord by means of chronically implanted macroelectrodes as described in detail in previous reports (Deliagina et al., 2000; Deliagina and Fagerstedt, 2000). In short, the electrodes (silver wires 75 μm in diameter and 3 mm in length) were oriented in parallel to the long spinal axons. They allowed an almost exclusive recording of the spike activity from larger fibres that have a conduction velocity of more than 2 m s^{-1}. In the lamprey, such a high conduction velocity is a characteristic of larger RS neurons (Rovainen, 1967, 1979). The electrodes were glued to a plastic plate (4 mm long, 2 mm wide and 0.25 mm thick). Two different designs of the electrode array were used, one with two electrodes and the other with four electrodes (Fig. 1A).

Surgery

Animals were operated on twice under MS-222 (Sigma-Aldrich, Stockholm, Sweden) anesthesia (100 mg l^{-1}). During the first surgery, the UL was performed. Sixty days after UL, when all animals reached a compensated state (that is they swam without rotation; see Deliagina, 1997a), the second surgery was performed and the recording electrodes were implanted.

The UL was performed either on the left side (N=3) or on the right side (N=3) using the technique described in detail previously (Deliagina, 1995, 1997a). In short, a hole was made in the dorsolateral aspect of the vestibular capsule and the labyrinth was removed with a pair of fine forceps under visual control. After removal, the intact medial wall of the vestibular capsule and a stump of the eighth nerve could be seen. *Post mortem* investigation showed that, in all cases, removal of the vestibular organ was complete and the medial wall of the capsule was undamaged.

The implantation of electrodes was performed as described in detail by Deliagina et al. (2000) and by Deliagina and Fagerstedt (2000). Two plates with electrodes were implanted at different rostrocaudal levels. The plate with two electrodes was implanted at the level of the third gill, and the plate with four electrodes 20–30 mm more caudally. The electrodes were facing the dorsal aspect of the spinal cord, as shown schematically in Fig. 1A.

Vestibular stimulation

Vestibular responses of RS neurons were recorded 1 or 2 days after implantation of the electrodes. The arrangements for vestibular stimulation, as well as the characteristics of stimuli, were described previously (Deliagina and Fagerstedt,

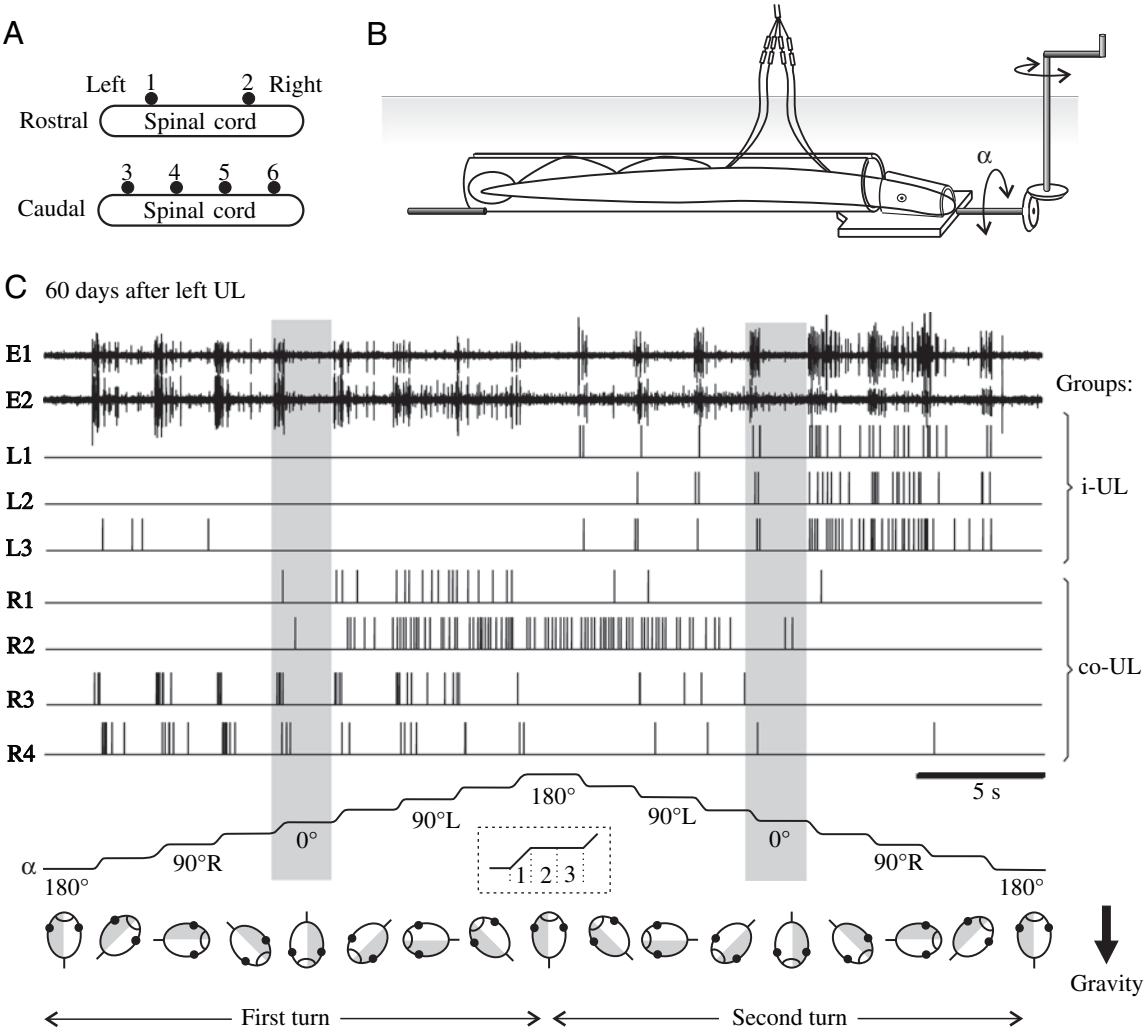

Fig. 1. (A) Positioning of electrodes for recording the activity in RS pathways. One array (electrodes 1 and 2) was implanted at the level of the third gill, and the other array (electrodes 3–6) 20–30 mm more caudally. (B) Arrangement for recording vestibular responses in RS neurons. The lamprey was positioned in the tube and rotated around its longitudinal axis (α, roll tilt angle). (C) A representative example of vestibular responses in RS pathways and individual RS axons in the animal compensated after the left unilateral labyrinthectomy (UL). Two sequential full turns (clockwise and counterclockwise) were performed in 45° steps. Shaded rectangles indicate the normal (horizontal) orientation of the animal. Positions of the animal in successive steps (in relation to the direction of gravity force) are shown. The left half of the lamprey body is shaded. Vestibular responses were measured separately for each of the three intervals of a step (inset); the activity in interval 1 (during rotation) will be considered as a dynamic response, the activity in intervals 2 and 3, as early and late static responses, respectively. Traces E1 and E2 show the mass activity in RS pathways recorded by the left and right electrodes of the rostral array, respectively (electrodes 1 and 2 in A). Seven neurons were separated from the mass activity using the spike-sorting program. The neurons L1–L3 had their axons located on the left, ipsilateral to the UL side of the spinal cord (i-UL group). The neurons R1-R4 had their axons located on the right, contralateral to the UL side (co-UL group).

2000). In short, the lamprey was positioned in the tube and rotated about the longitudinal body axis (Fig. 1B). To reveal the dynamic characteristics of vestibular responses, they were rotated in 45° steps. To reveal a directional sensitivity of neurons, two full turns were produced in opposite directions (Fig. 1C, bottom). To examine the effect of tonic visual input on vestibular responses, the responses were examined both in light and in darkness. Testing in light was performed in an aquarium that was illuminated by a 100 W white incandescent lamp mounted above the aquarium at a distance of 2 m. A considerable part of the light was reflected from a sheet of white paper positioned under the transparent bottom of the aquarium, thus producing a rather diffuse illumination within the aquarium.

Data processing

Signals from the electrodes were amplified by conventional AC amplifiers, digitized with a sampling frequency of 10 kHz and stored on the hard disk of an IBM AT compatible computer by means of data acquisition software (Digitdata 1200/Axoscope, Axon Instruments, Inc., Union City, CA, USA). The recorded multiunit spike trains were separated into unitary

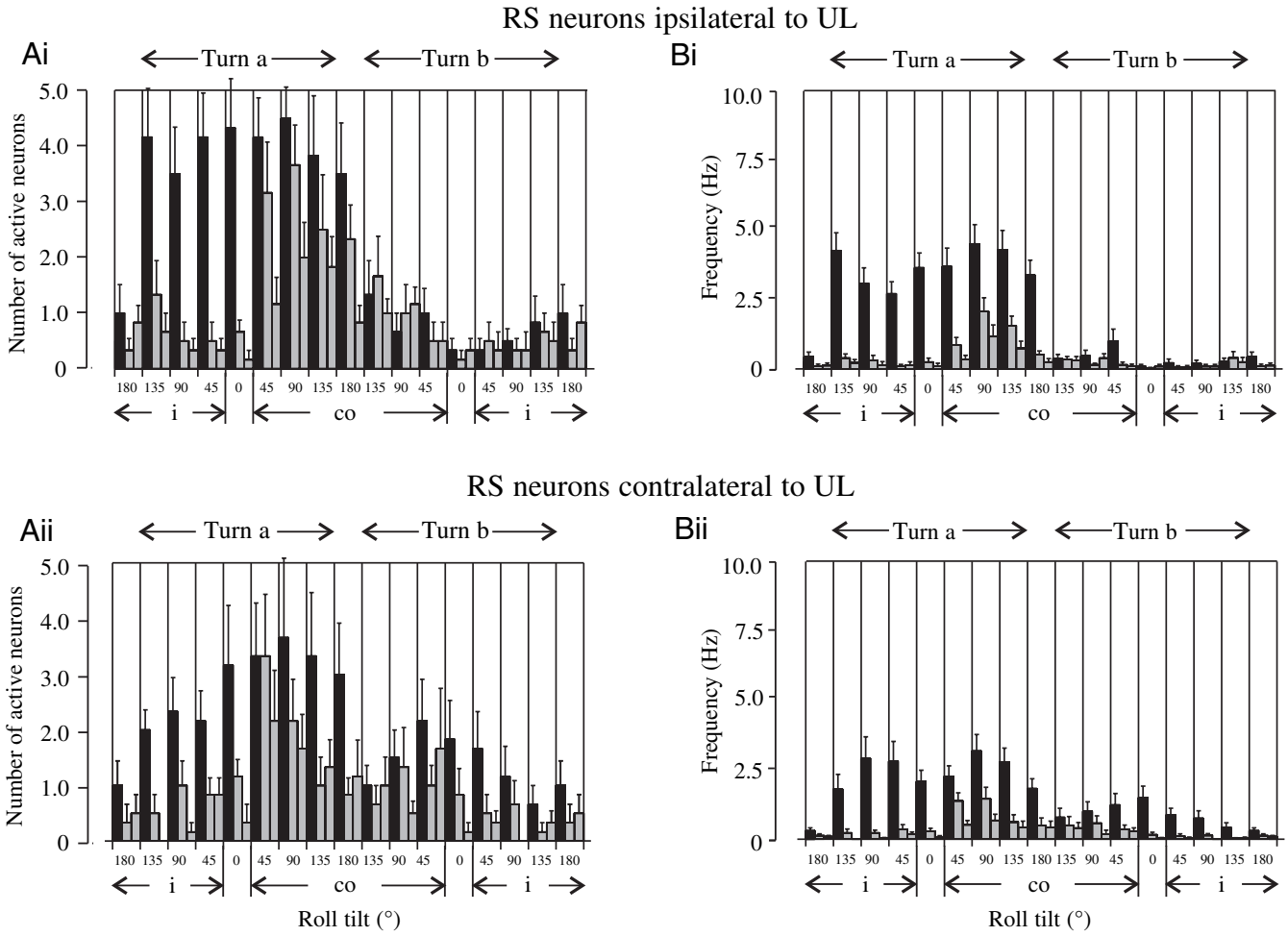

Fig. 2. Summary of responsesto full turn rotation in RS neurons of compensated UL-animals, tested in light. (Ai,Aii) Number of active neurons as a function of the roll angle. This value was calculated as the number of neurons activated in each angular step in each of the animals, and then averaged over all six animals. (Bi,Bii) Average discharge frequency of neurons as a function of roll angle. This value was calculated as the number of spikes per second generated by each neuron in each step and then averaged over all active neurons in the group (N=31 and 30 for Bi and Bii, respectively). The 0° angle corresponds to the dorsal-side-up orientation of the lamprey. The angular zones where the ipsilateral (i) or contralateral (co) labyrinth was facing downward are indicated. Rotation was performed towards the contralateral labyrinth in turn a, and towards the ipsilateral one in turn b. Each step of rotation was divided into three intervals, and responses were calculated separately for each interval (see inset and legend in Fig. 1). In each of the steps, the dynamic response (activity during rotation) is shown by a black bar, the early and late static responses are shown by two successive shaded bars. Values are means ± S.E.M.

waveforms, representing the activity of individual axons, by means of data analysis software (‘Spike sorting’, Datapac III, Run Technologies, Inc., Laguna Hills, CA, USA). The analysis was based on the selection of distinguishable unitary waveforms occurring on one electrode, or occurring simultaneously on two or more electrodes of the array; this technique was previously described in detail (Deliagina and Fagerstedt, 2000; Deliagina and Pavlova, 2002; Pavlova and Deliagina, 2002a, 2003).

To determine the angular zones of sensitivity of individual RS neurons, their vestibular responses were characterized quantitatively. For this purpose, each step of rotation was divided into three intervals (1–3, see inset in Fig. 1C), and the firing frequency of a neuron was measured separately for each interval in each step. The activity in interval 1 (during movement) will be termed ‘dynamic response’; the activity in intervals 2 and 3 (when a new position was maintained) will be termed ‘early’ and ‘late static responses’, respectively.

The mediolateral position of individual axons in the spinal cord was estimated by comparing the amplitudes of the same spike recorded by different electrodes of the same array. The conduction velocity in individual axons could also be measured using the time delay between spikes from the same axon recorded by the rostral and caudal electrodes (Deliagina and Fagerstedt, 2000).

All analytical procedures and possible sources of errors during the spike sorting have also been fully described in recent reports (Deliagina and Fagerstedt, 2000; Deliagina and Pavlova, 2002; Pavlova and Deliagina, 2002a, 2003).

All quantitative data in this study are presented as the mean

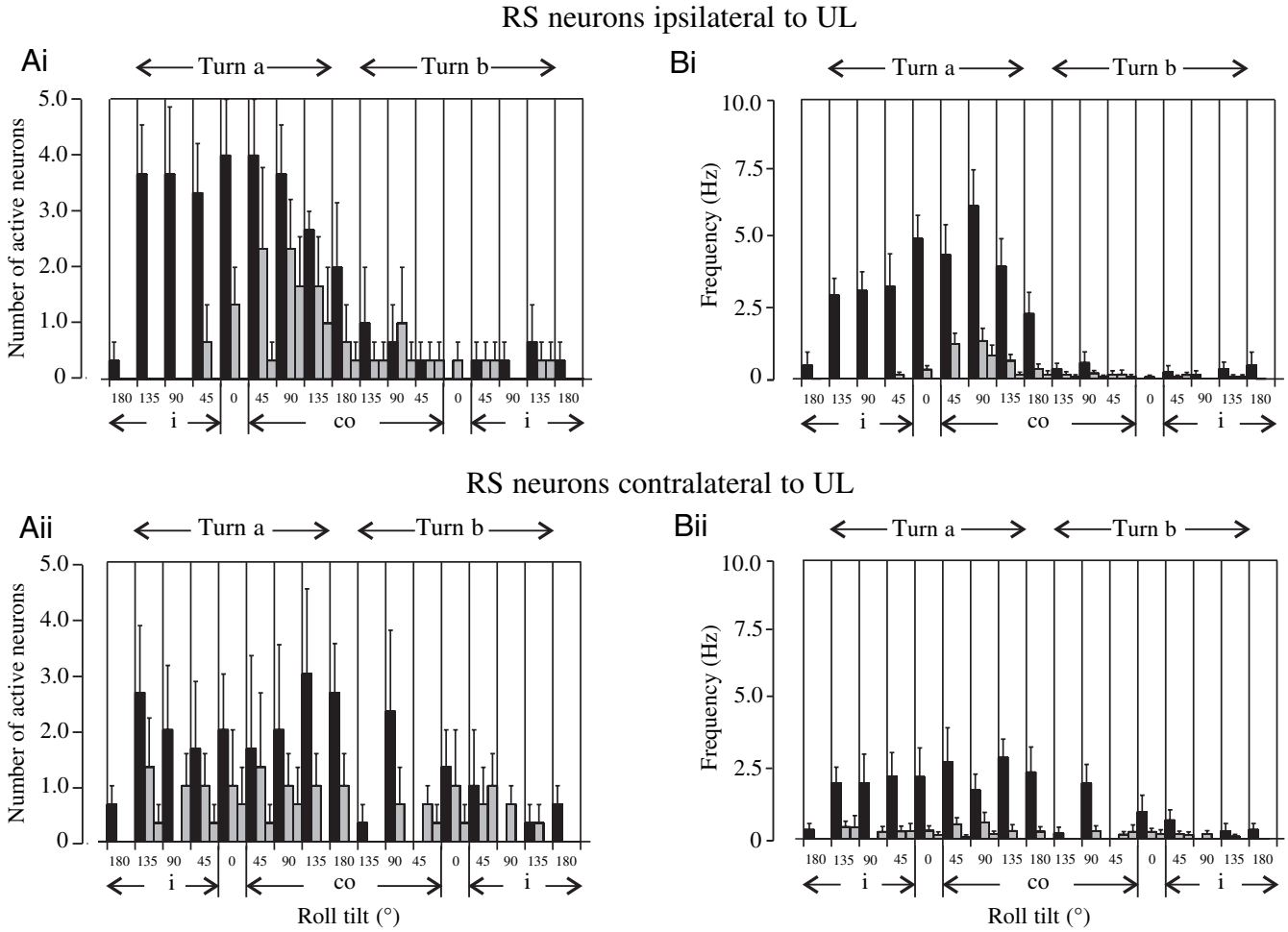

Fig. 3. Summary of responses to full turn rotation in RS neurons of compensated UL-animals tested in darkness. (Ai,Aii) Number of active neurons as a function of roll angle. (Bi,Bii) Average discharge frequency of neurons as a function of roll angle (N=14 and 12 for Bi and Bii, respectively). Designations as in Fig. 2.

± S.E.M. Paired Student's t-tests were used to determine the statistical significance when comparing different means; the confidence level was set at P=0.05. All statements in the following text about the similarity or difference between the neuronal responses are based on these statistical criteria.

Results

From seven to 14 neurons were recorded in individual animals. For the cases when an axon was recorded by both rostral and caudal electrodes (28% of all neurons tested), the conduction velocity was calculated. In all cases the spikes propagated in the rostrocaudal direction. The velocity ranged from 2.4 to 4.4 m s^{-1}. In lampreys, no descending axons with such a high conduction velocity, besides the RS ones, have been reported.

Vestibular responses of RS neurons in light

The activity in RS pathways in light was recorded in all six compensated UL animals. Normally, the resting activity in RS neurons was low or absent, and vestibular stimulation activated the neurons. This is illustrated in Fig. 1C for the animal that was fully compensated after the left UL. Traces E1 and E2 show the mass activity in RS pathways recorded by the left and right electrodes of the rostral array, respectively (electrodes 1 and 2 in Fig. 1A). Using the spike-sorting program, the activity of seven individual axons was separated from the mass activity. All three neurons with their axons located on the left side of the spinal cord, that is, ipsilateral to the UL (L1–L3, the i-UL group) had very similar patterns of responses. In the first turn (rotation toward the UL) they exhibited almost no activity. In the second turn (rotation toward the intact, contralateral labyrinth), the neurons exhibited a dynamic response with any change of position. In addition, they had static responses within the zone 45°R to 135°R.

The neurons with their axons located on the right side of the spinal cord (R1–R4, the co-UL group) exhibited more diverse patterns of responses. The neurons R1 and R2 responded statically at the positions when the UL side was facing downward, either in the first turn (R1) or in both turns (R2). The neurons R3 and R4 responded dynamically to any change

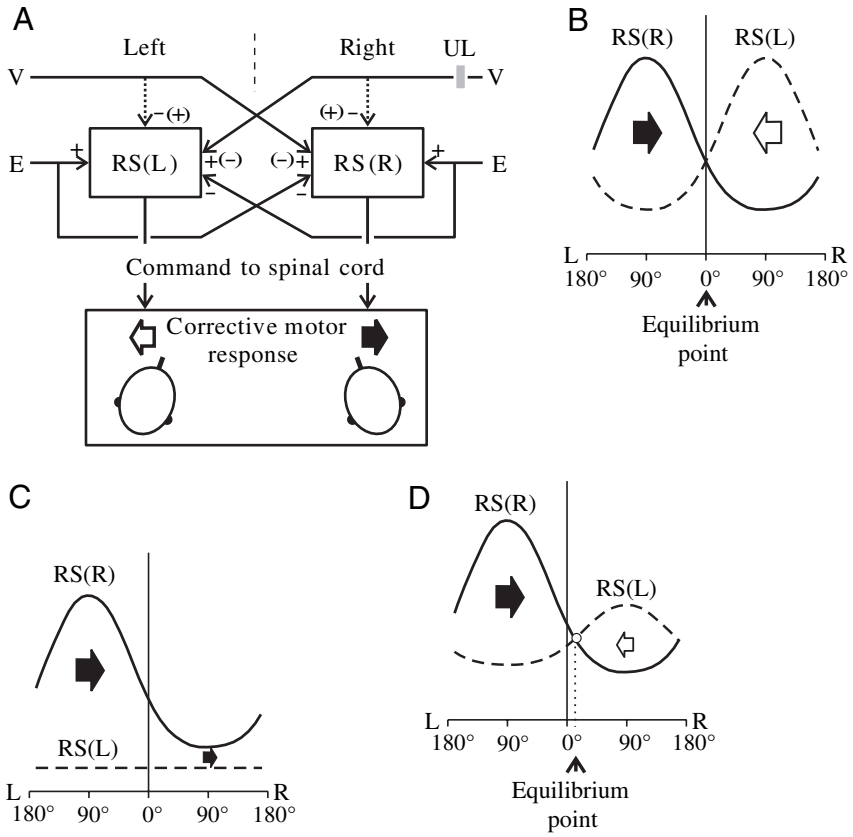

Fig. 4. Schematic outline of the impairment of the postural system caused by UL, and restoration of postural function in compensated lampreys. (A) Conceptual model of the roll control system as proposed previously (Deliagina and Pavlova, 2002). Left and right groups of RS neurons, RS(L) and RS(R), receive vestibular (V) and visual (E) sensory inputs. The plus and minus signs without parentheses indicate the major effects on RS neurons produced by these inputs, the signs in parentheses indicate the minor effects. Weaker inputs from the ipsilateral labyrinths are shown by dotted lines. The presumed directions of rolling caused by RS(L) and RS(R) are indicated by the white and black arrows, respectively. (B–D) Operation of the model under different conditions. The curves represent the activity of the left and right groups of RS neurons as a function of roll angle. (B) Control (intact lamprey). The two activity curves intersect, and the system has an equilibrium point at 0° (dorsal-side-up orientation). (C) The right labyrinth removed (shown by a shaded rectangle in A). The system has no equilibrium point. (D) The result of vestibular compensation. Plastic changes in the postural network caused a restoration of vestibular responses in RS(L) neurons (as a result of augmentation of input from the ipsilateral labyrinth) and recreation of the equilibrium point.

of position in the first turn, whereas their static responses were weak (in R3) or absent (in R4).

Altogether, 64 RS neurons were recorded in six animals in the light. The overwhelming majority of them (61 neurons, or 95%) exhibited specific responses to vestibular stimulation: they were activated more strongly by rotation towards the contralateral labyrinth than in the opposite direction and/or they had specific angular zones of responses. A small proportion of neurons (3 units, or 5%), were activated by tilts in any direction, and their activity did not correlate with any particular spatial orientation. In intact animals, such neurons were also rarely observed (Deliagina and Fagerstedt, 2000). Of the 61 neurons with specific vestibular responses, 31 neurons were located on the side ipsilateral to the UL (the i-UL group) and 30 neurons on the opposite side (the co-UL group).

To describe qualitatively the vestibular responses in the i-UL and co-UL neuron groups, two characteristics were used (Deliagina and Fagerstedt, 2000; Deliagina and Pavlova, 2002; Pavlova and Deliagina, 2002a, 2003): (1) the number of simultaneously active neurons, and (2) the mean discharge frequency of these neurons. The number of active neurons was calculated separately for each animal and then averaged over all six animals.

Fig. 2Ai shows a histogram of the number of simultaneously active i-UL neurons. Along the horizontal axis, the successive angles of roll tilt during two turns (a and b) performed in opposite directions are indicated. To combine the data obtained with the left and right UL, turn a represents the responses obtained with rotation toward the intact labyrinth, and turn b, toward the UL. One can see that during turn a any change of orientation evoked dynamic responses in most neurons. During turn b, the dynamic responses were much weaker than in turn a. Static responses were also more pronounced in turn a, at the positions $45°_{co}$ to $135°_{co}$. When the same positions were reached by rotation in the opposite direction (turn b), only a small proportion of neurons were statically activated.

To evaluate the directional sensitivity of i-UL neurons, for each of the animals we calculated the mean number of neurons responding dynamically to sequential steps in turn a, and then averaged this value over all six animals; similar calculations were performed for turn b. The mean value of response in turn a was 3.6±0.1 neurons *versus* 0.8±0.1 neurons in turn b. The difference was statistically significant.

Fig. 2Bi shows the frequency curve for the i-UL group of RS neurons. One can see that the neurons were active mostly in turn a. In this turn, the dynamic responses were much stronger than the static ones. The mean value of the dynamic responses in turn a was 3.3±0.4 Hz *versus* 0.4±0.1 Hz in turn b. This difference was also statistically significant.

Thus, according to both characteristics (the number of active neurons and their frequencies), the principal feature of the i-UL neurons is their much stronger responses in turn a as compared to turn b. In this respect, the responses were similar to those of RS neurons in intact lampreys observed in the previous studies (Deliagina and Fagerstedt, 2000; Deliagina and Pavlova, 2002).

As shown previously (Deliagina and Pavlova, 2002), the main effect of UL in the non-compensated animals was the lack of activity (absence of vestibular responses) in co-UL neurons (see figs 5B2 and 6B2 in Deliagina and Pavlova, 2002). In contrast, the present study showed that this population in the compensated animals was active. In six animals, we recorded 30 co-UL neurons responding to vestibular stimulation. This number was almost equal to the number of i-UL neurons (N=31) recorded by the same electrodes. Fig. 2Aii shows a histogram of the number of simultaneously active co-UL neurons, and Fig. 2Bii, a histogram of their frequencies. Turn a is the responses obtained with rotation toward the UL, and turn b with rotation toward the intact labyrinth. Both histograms were qualitatively similar to those for i-UL neurons (Fig. 2Ai,Bi), that is, the responses in turn a were larger than the responses in turn b. For the number of active neurons, the mean value of responses in turn a was 2.6±0.4 neurons versus 1.4±0.2 neurons in *turn b*. For the frequency curve, the mean value of responses in turn a was 2.2±0.3 Hz *versus* 0.9±0.1 Hz in turn b. The difference in both cases was statistically significant. However, the response magnitude in turn a in the co-UL group was slightly smaller than in the i-UL group (compare Fig. 2Bii,Bi). The mean value of responses in the co-UL group was 2.2±0.3 Hz versus 3.3±0.4 Hz in the i-UL group. The difference was statistically significant.

Vestibular responses of RS neurons in darkness

To characterize the significance of visual input for the generation of vestibular responses in the compensated animals, three animals (out of six animals tested in light, see above) were also tested in darkness. The main result of these tests was that vestibular responses on both sides persisted in darkness. In the i-UL group, the responses in darkness (Fig. 3Ai,Bi) were very similar to those in light (Fig. 2Ai,Bi). In the co-UL group, the response pattern in darkness (Fig. 3Aii,Bii) was also similar to that in light (Fig. 2Aii,Bii). However, a total number of responding i-UL and co-UL neurons in the three animals tested in darkness (N=14 and 12, respectively) was smaller than in the same animals tested in light (N=21 and 18, respectively). The difference was statistically significant.

Discussion

Since the postural control system in the lamprey is driven almost exclusively by vestibular input, the effect of a unilateral labyrinthectomy (UL) in this animal is dramatic – it results in a complete loss of equilibrium and continuous rolling when swimming (de Burlet and Versteegh, 1930; Ullén et al., 1995a; Deliagina, 1995, 1997a). The recovery of equilibrium control, which is an essential component of vestibular compensation, is relatively slow and usually takes a few weeks (Deliagina, 1995, 1997a); this is a much longer time than, for example in rats (less than one day; Deliagina et al., 1997).

The key elements of the postural system in the lamprey are the left and right groups of reticulospinal (RS) neurons, transmitting commands for postural corrections to the spinal cord (Deliagina et al., 2002). Recently it was found that UL causes a dramatic asymmetry in these commands: the vestibular-evoked activity on the lesioned side persisted after UL, whereas the activity on the opposite side disappeared completely (Deliagina and Pavlova, 2002). It was also found that illumination of the eye contralateral to the UL results in a restoration of symmetry in the bilateral activity of RS system. Since illumination of this eye also leads to a restoration of equilibrium in swimming, non-compensated lampreys (Deliagina, 1997b), it was suggested that the loss of equilibrium in UL lampreys is caused by the asymmetry in the descending RS commands, and a recovery of postural control during a process of vestibular compensation is the result of a restoration of symmetry in these commands.

The main result of the present study is that vestibular responses in RS neurons on the side contralateral to the UL (the co-UL group), which were absent when tested in the non-compensated animals a few days after UL (Deliagina and Pavlova, 2002), were present in the compensated animals (Fig. 2Aii,Bii). This finding strongly supports the hypothesis that a restoration of bilateral symmetry in the RS commands underlies a recovery of equilibrium control.

However, the restored vestibular responses in the co-UL neurons (Fig. 2Aii,Bii) differed, to some extent, from the normal ones in intact animals (Deliagina and Pavlova, 2002), or from the responses in the i-UL group with the main (contralateral) vestibular input intact (Fig. 2Ai,2Bi). First, the magnitude of the restored responses was reduced by 30–40%. Second, the co-UL group was less homogenous than the i-UL group: in addition to the neurons responding mainly in the turn towards the UL, some neurons responded in both turns (Fig. 1C).

In behavioral experiments it was found that the process of vestibular compensation in lampreys strongly depends on the presence of visual input. Upon reaching a compensated state, however, this input becomes nonessential for the maintenance of equilibrium, and the eyes can even be removed (Deliagina, 1997a,b). It was suggested that in the compensated animals the restored activity in the co-UL neurons does not require any significant support from the visual input. This hypothesis was confirmed in the present study; we found that vestibular responses in the co-UL neurons that were observed in light (Fig. 2Aii,Bii), persisted also in darkness (Fig. 3Aii,Bii), though their magnitude was slightly reduced. These data also support a more general assumption that restoration of the 'central symmetry' (i.e. the symmetry in the activity of vestibular nuclei and their targets) constitutes an essential component of vestibular compensation (Deliagina et al., 1997; for a review, see Curthoys and Halmaguyi, 1999).

A possible functional role of the restored activity in the co-UL neurons can be considered in the framework of the conceptual model of the roll control system proposed earlier (Deliagina, 1997a; Deliagina and Pavlova, 2002; Zelenin et al., 2000) (Fig. 4A). The key elements of the model are the left and right groups of RS neurons, RS(L) and RS(R). The main input

to these neurons is from the contralateral labyrinth, whereas the input from the ipsilateral labyrinth is much weaker. Each input contains both excitatory and inhibitory components; the components depend differently on the tilt angle (Deliagina and Pavlova, 2000). Owing to these inputs, the activity of RS neurons is orientation dependent, with its peak at ~90° of contralateral roll tilt (Fig. 4B). The two groups of neurons also receive an excitatory input from the ipsilateral eye and an inhibitory input from the contralateral eye. It was suggested that each of the groups, via spinal mechanisms, elicits ipsilateral rotation of the animal (black and white arrows in Fig. 4A,B). The system will stabilize the orientation in space with equal effects produced by RS(L) and RS(R), that is, the dorsal-side-up position (equilibrium point in Fig. 4B).

The model explains the loss of equilibrium after UL in the following way. As a result of the loss of the main vestibular input from the contralateral labyrinth, the UL causes inactivation of RS neurons on the contralateral side (Deliagina and Pavlova, 2002), as illustrated for the right-side labyrinthectomy in Fig. 4C. Because of the inactivation of RS(L), the two activity curves no longer intersect, the system has no equilibrium point, and the dominating RS(R) causes the main postural deficit: rolling of the lamprey to the right.

The model implies that restoration of postural equilibrium during vestibular compensation is due to a recovery of activity in the co-UL group of RS neurons, so that this group of neurons can counteract the i-UL group, and the two activity curves will intersect again (Fig. 4D). The present study has shown that the vestibular-induced activity indeed re-appeared in the co-UL neurons (Fig. 2Aii,Bii and Fig. 3Aii,Bii). The angle at which the RS(L) and RS(R) curves will intersect depends on the degree of restoration of the activity (vestibular responses) in the deafferented RS neurons. If these responses are weaker than those in the i-UL neurons, the equilibrium point will be shifted toward the UL (as in Fig. 4D), and the roll control system will stabilize this tilted position – the behavior often observed in the compensated animals (Deliagina, 1997a).

Presumed cellular and network mechanisms underlying the recovery of central symmetry after UL in amphibians and mammals have been discussed by a number of authors (Darlington and Smith, 1996; Ris et al., 1995, 2001; Vibert et al., 1999; Curthoys and Halmagyi, 1999; Smith and Curthoys, 1989; Dieringer, 1995). Some of these mechanisms can be considered in relation to the lamprey.

In previous studies it was shown that two inputs to RS neurons, from the contralateral and ipsilateral labyrinths (Fig. 4A), have similar spatial zones of sensitivity and thus supplement each other when eliciting vestibular responses in RS neurons. The ipsilateral input, however, is much weaker than the contralateral one and, when acting alone, is not able to activate RS neurons in the non-compensated animals (Deliagina and Pavlova, 2002). The present study has shown that recovery of equilibrium in UL animals is accompanied by the appearance of responses of RS neurons to the signals coming from the ipsilateral labyrinth. The appearance of responses to previously sub-threshold signals can be explained by changes in the membrane properties (excitability) of either RS neurons themselves, or pre-reticular neurons transmitting vestibular signals, as well as by changes in synaptic efficacy of the existing synapses and/or reactive synaptogenesis in the vestibulo-reticular pathways. An increase of the tonic excitatory drive to RS neurons from other sources can also contribute. These factors could explain the appearance of responses within the angular zones similar to the normal ones. However, an increased diversity of angular zones of restored responses as compared to normal responses (Fig. 1C) suggests the appearance of new connections originating from the vestibular afferents with corresponding characteristics of their spatial sensitivity.

In conclusion, the present study has demonstrated that, in the lampreys subjected to ablation of one labyrinth, the recovery of an important motor function – the maintenance of equilibrium – is associated with a restoration of a close-to-normal pattern of supraspinal motor commands. The corresponding plastic changes in brainstem neuronal networks remain to be identified.

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