Monosynaptic excitation of trochlear motoneurons following electrical stimulation of the prepositus hypoglossi nucleus

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Recent evidence indicates that the prepositus hypoglossi nucleus subserves a role in eye movements. Morphological studies by Graybiel and Hartweg suggest that prepositus neurons might terminate within the oculomotor nucleus. The purpose of the present study was to determine whether stimulation of the prepositus hypoglossi nucleus could produce synaptic potentials in trochlear motoneurons and, if so, to ascertain the excitatory and/or inhibitory nature of the projection.

As a result of the intimate relationships of the prepositus nucleus with the medial vestibular, abducens and underlying reticular nuclei electrophysiological data obtained with a central stimulating electrode must be evaluated with caution. Despite this reservation, it is possible to report that there is a modest ipsilateral excitatory projection from the prepositus nucleus to trochlear and all other oculomotor neurons. The evidence supporting this contention has been selected from three experiments in the trochlear nucleus in deference to its isolated midbrain localization and well-studied synaptic organization.

Cats were anesthetized with pentobarbital (30 mg/kg) paralyzed with Flaxedil and artificially respired. Bipolar stimulating silver ball electrodes were placed on each VIIIth nerve extracranially (Fig. 1A). Concentric bipolar stimulating electrodes were positioned near the anterior part of the prepositus hypoglossi nucleus (Fig. 1A) juxtaposed to the medial longitudinal fasciculus in order to recruit maximum nuclear output. In the experiments with both acute and chronic brain stem lesions cerebellar lobules IX and X were removed to facilitate separation of the vestibular nucleus complex from the prepositus hypoglossi nucleus (Fig. 1A). A detailed reconstruction of these lesions will be presented subsequently; however, their adequacy was shown physiologically by the absence of characteristic vestibular fields in the trochlear nucleus following vestibular stimulation (Fig. 1B). Trochlear motoneurons were antidromically identified following electrical stimulation of the IVth nerve in the orbit.

The field potential profiles recorded near the center of trochlear nucleus following electrical stimulation of ipsilateral prepositus (iPH), trochlear nucleus (Tro), contralateral excitatory (Vc) and ipsilateral inhibitory (Vi) pathways are shown by the
upper records in Fig. 1B. The ipsilateral inhibitory (Vi) and contralateral excitatory (Vc) pathway are characteristically positive and negative respectively.

Following iPH stimulation (up to 5 times threshold) one observes a sharp negativity followed by a slower negativity (arrows). The latter potential often exhibits two clear deflections. A similarly shaped iPH field potential profile can be recorded throughout the oculomotor complex, some lateral regions of the midbrain and areas in the superior colliculus. Due to this spatial spread of the iPH field it is not possible to assume that the two negativities represent the pre- and post-synaptic current underlying synaptic potentials in trochlear motoneurons; however, this is likely as shown by the intracellular short latency depolarization (EPSPs) following iPH (2 times threshold) stimulation (Fig. 1C). The latency for this depolarization was about 0.7 msec indicating its monosynaptic nature. Stimulation of the contralateral prepositus (cPH) produces similar extra- and intracellular potentials with a slightly longer latency (0.1 msec) but much smaller amplitudes at comparable stimulus intensities. Based on other experiments with electrical stimulation of the iPH and cPH we feel the ascending pathway to originate, primarily, from the ipsilateral prepositus hypoglossi nucleus.

Due to the intimate relationship of the prepositus nucleus with the vestibular complex as well as the proximity of the iPH stimulating electrode to the vestibular complex, inhibitory and excitatory vestibular neurons were activated (on occasion) by prepositus stimulation. This was revealed by axonal recordings in the trochlear nucleus (Fig. 2D and E). In addition, as shown in acute experiments, IPSPs were elicited in trochlear motoneurons with a latency of about 1 msec (arrows in B and C).
Fig. 2. Intracellular records from trochlear motoneurons and axons of secondary vestibular neurons subsequent to prepositus stimulation. A–C: intracellular responses from an identified Tro Mn. Control synaptic potentials are shown in A following iPH, Vc and Vi stimulation respectively. Responses in B were obtained immediately after injecting Cl⁻ (10⁻⁸ A for 2 min) from a NaCl micropipette. In C the synaptic potentials were taken during DC application of 2 × 10⁻⁸ A depolarizing current. D and E: intracellular axonal records from an inhibitory (upper) and excitatory (lower) vestibular neuron projecting to the trochlear nucleus. In D, iPH stimulation was at 2.5 times threshold (straddling intensity) and in E at a suprathreshold stimulation intensity (4 times threshold).

Following isolation of the prepositus hypoglossi nucleus by bilateral parasagittal lesion (Fig. 1A) the Vi positivity and Vc negativity were significantly reduced in the trochlear nucleus (compare control (Con) and lesion (Les)); however, the iPH field potential profiles were retained. At that time it was no longer possible to evoke inhibition (checked with Cl⁻) following iPH stimulation even though monosynaptic EPSPs were still elicited in trochlear motoneurons (Fig. 3A). After chronic parasagittal lesions (Fig. 3B) it was still possible to record short latency depolarization (EPSPs) in trochlear motoneurons (Tro Mns) following iPH stimulation. Moreover, the latency for the EPSPs in the chronic experiments (1.0–1.2 msec) was significantly longer (by

Fig. 3. Depolarization in trochlear motoneurons following acute and chronic parasagittal lesions isolating the prepositus from the vestibular nuclei. A: synaptic depolarization (latency 0.7 msec) recorded in a trochlear motoneuron following the acute parasagittal lesions shown in Fig. 1A (same experiment as in 1B). In the upper records of B the EPSPs are shown in an antidromically identified Tro Mn 5 days after parasagittal lesions indicated in Fig. 1A. The lower extracellular record is 2.5 times the AC gain of the upper one in order to facilitate comparison of the field potential profile from the chronic with that of normal preparation shown in Fig. 1B.
about 0.5 msec) than in normal acute experiments. This suggests that the short latency depolarization in the acute experiments was probably due to axon collateral activation of secondary excitatory vestibular neurons (Fig. 1A).

These results demonstrate a unique excitatory path from the iPH area to Tro Msn distinct from the vestibular excitatory projection. Furthermore, the apparent absence of inhibition following either acute or chronic vestibular prepositus separation indicates that the IPSPs found in the acute physiological situations are likely to have been produced by activation of axon collaterals from inhibitory vestibular neurons.

Overall, the data indicate a modest, monosynaptic excitatory projection from the ipsilateral prepositus hypoglossi to trochlear motoneurons. This conclusion is consistent to date with our findings in other vertical oculomotor neurons; however, chronic (in this case control) experiments excluding complicating axon collateral effects are required before the extent of the complete prepositus projection may be stated with certainty.

Since the prepositus projection to trochlear motoneurons appears to be mainly ipsilateral and solely excitatory in nature it thereby is organized quite differently from the reciprocal inhibitory–excitatory vestibular ocular projection. The relatively weak excitatory synaptic connections directly with trochlear motoneurons if true for other oculomotor neurons, is in contrast to the strong vestibuloocular connections. These observations do not preclude a significant role for the prepositus nucleus in oculomotor organization because this posterior brain stem area may receive and exert direct effects on other vestibular–reticulo–cerebello–ocular nuclei. Such a widespread influence might be summated to the point where it provides a critical regulatory function in eye movement.

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