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SEGREGATION OF ELECTRO- AND MECHANORECEPTIVE INPUTS TO THE ELASMOBRANCH MEDULLA

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SUMMARY

The anterior lateral line nerve of the thornback ray consists of fibers that innervate head electroreceptive ampullary organs and mechanoreceptive neuromasts. As the anterior lateral line nerve enters the medulla it divides into dorsal and ventral roots. Single unit responses of dorsal root fibers to electric field and mechanical stimuli indicate that the dorsal root consists only of ampullary fibers, whereas the ventral root consists only of mechanoreceptive fibers. The dorsal and ventral roots of the anterior lateral line nerve terminate in the dorsal and medial octavolateralis nuclei respectively, indicating that the dorsal nucleus is the primary electroreceptive nucleus of the elasmobranch medulla and the medial nucleus is the mechanoreceptive nucleus. Averaged evoked potential responses to electric field stimuli could be recorded from the dorsal but not the medial nucleus, further evidence that the dorsal nucleus is the electroreceptive nucleus. A second evoked response to electric field stimuli was elicited from the lateral reticular nucleus, suggesting that the reticular formation may be a secondary target of efferents of the dorsal octavolateralis nucleus. A dorsal octavolateralis nucleus exists not only in elasmobranchs, but also in agnathan, chondrostean, dipnoan, and crossopterygian fishes, suggesting that all of these taxa are also electroreceptive.

INTRODUCTION

The lateral line system of cartilaginous fishes consists of at least two peripheral

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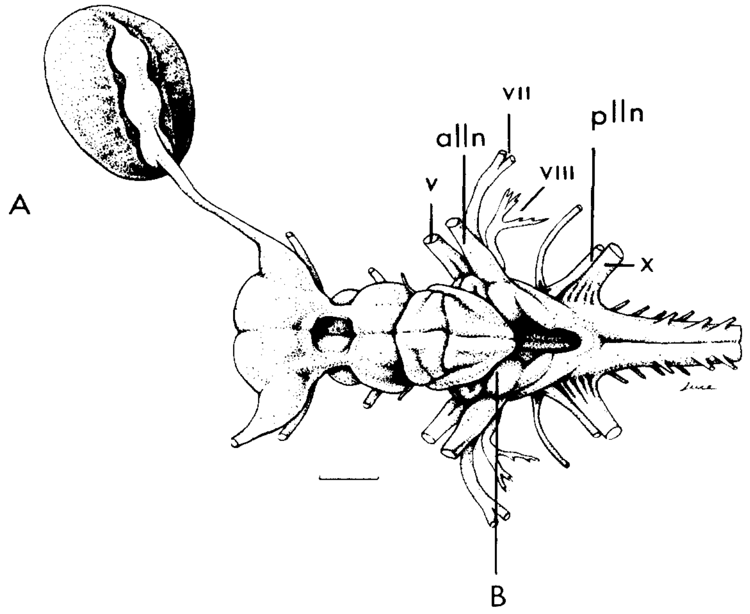

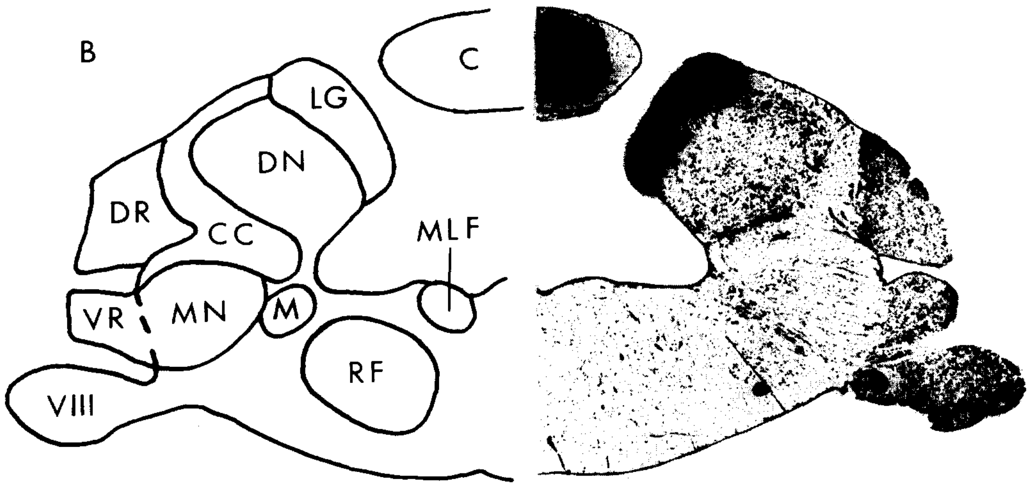

Fig. 1. A: dorsal view of the brain of the thornback ray. Bar scale equals 5 mm. B: transverse section through the medulla illustrating the positions of the dorsal and medial octavolateralis nuclei. Abbreviations: alln, anterior lateral line nerve; B, position of transverse section in medulla; C, corpus of cerebellum; CC, cerebellar crest; DN, dorsal octavolateralis nucleus; DR, dorsal root of anterior lateral line nerve; LG, lateral granule cell mass; M, magnocellular octavus nucleus; MLF, medial longitudinal fasciculus; MN, medial octavolateralis nucleus; plln, posterior lateral line nerve; RF, reticular formation; VR, ventral root of anterior lateral line nerve; V, trigeminal nerve; VII, facial nerve; VIII, octavial nerve; X, vagal nerve.

receptor types: mechanoreceptive neuromasts and electroreceptive ampullary organs[7,9]. Ampullary organs are restricted to the head in sharks but are also found on the pectoral fins in skates and rays[6,13]. In all cartilaginous fishes, ampullary organs and head neuromasts are innervated by the anterior lateral line nerves, whereas the trunk neuromasts are innervated by the posterior lateral line nerves.

Recently, Boord and coworkers[3,10,12] experimentally traced the central course of the lateral line nerves in sharks and skates. Both the anterior and posterior lateral line nerves enter the octavolateralis medullar area, which consists of dorsal (anterior lateral line lobe), medial or intermedial (posterior lateral line lobe) and ventral nuclei (Fig. 1). As the anterior lateral line nerve approaches the medulla, it divides into dorsal and ventral roots (Fig. 1B) with the dorsal root terminating in the dorsal nucleus and the ventral root terminating in the medial nucleus[10,12]. The posterior lateral line nerve enters only the medial nucleus. Based on this anatomical arrangement, Boord and Campbell[3] suggested that ampullary fibers are restricted to the dorsal root of the anterior lateral line nerve, that the dorsal nucleus is thus the primary electroreceptive medullary nucleus, and that the medial nucleus is the primary mechanoreceptive nucleus receiving neuromast information from both head and trunk.

In order to test this hypothesis, we examined the sensitivity of single units of the dorsal and ventral roots of the anterior lateral line nerve in the thornback ray (*Platyrhinoidis triseriata*) to weak mechanical and electric field stimuli. In addition, averaged evoked potential responses to electric fields were recorded from the octavolateralis nuclei of the medulla.

MATERIALS AND METHODS

The medulla and lateral line nerves of 11 thornback rays (200–400 g) were exposed under tricaine methanesulfonate anesthesia (MS222, about 0.01 %). Animals were then paralyzed with tubocurarine chloride (5 mg/kg, i.v.) and placed in fresh sea water in a plexiglass aquarium (53 × 97 × 15 cm) considerably longer than the rays; only the dorsal surface of the head and exposed brain were above the water level. Oxygenated sea water (20–22 °C) flowed through a tube placed in one spiracle for ventilation.

Single unit responses were recorded using glass micropipettes of 5–20 MΩ resistance (4 M NaCl). Cell responses in some cases were analyzed using post-stimulus–time histograms of impulses, but most responses were readily apparent from single stimulus presentations. Evoked potential recordings were made with stainless steel electrodes of 1–5 MΩ resistance. Recording sites were localized by the Prussian blue technique[8].

Electric field stimuli were presented as DC pulses of 5–100 msec duration between pairs of 20 cm carbon rod electrodes on the sides or ends of the aquarium, thereby producing homogeneous fields with voltage gradients perpendicular or parallel to the longitudinal axis of the fish. Voltage pulses were supplied by a DC stimulator and isolation unit, and a series resistance of 10^5–10^6 Ω was used to reduce polarization effects. The voltage gradient resulting from this stimulus arrangement approximated uniformity over the length of the fish, and field intensities were monitored with a pair of silver wire electrodes in the bath near the fish.

Mechanical stimuli included weak substrate vibrations, stroking of the body surface with a brush, and disturbances in the water caused by water drops or striking the surface with a blunt probe.

RESULTS

All of the 82 units recorded in the dorsal root of the anterior lateral line nerve were responsive to weak electric fields, most with thresholds below 10 μV/cm (0.40 μA/sq.cm); 26 of the cells responded to stimuli less than or equal to 1 μV/cm (0.04 μA/sq.cm). Most units had regular ongoing activity, typically 20–35 imp./sec; but in very lightly curarized animals, cells produced bursts of spikes correlated with the animal's respiratory movements. Units typically had a best field orientation (parallel or perpendicular), and with the onset of a stimulus the impulse frequency sharply increased or decreased depending on the field polarity. With a given polarity the opposite effect on spike frequency was observed at stimulus offset (Fig. 2). Of the 82 units recorded, 34 were held for a sufficient length of time to test carefully for mechanosensitivity. The dorsal root units were unresponsive to weak mechanical

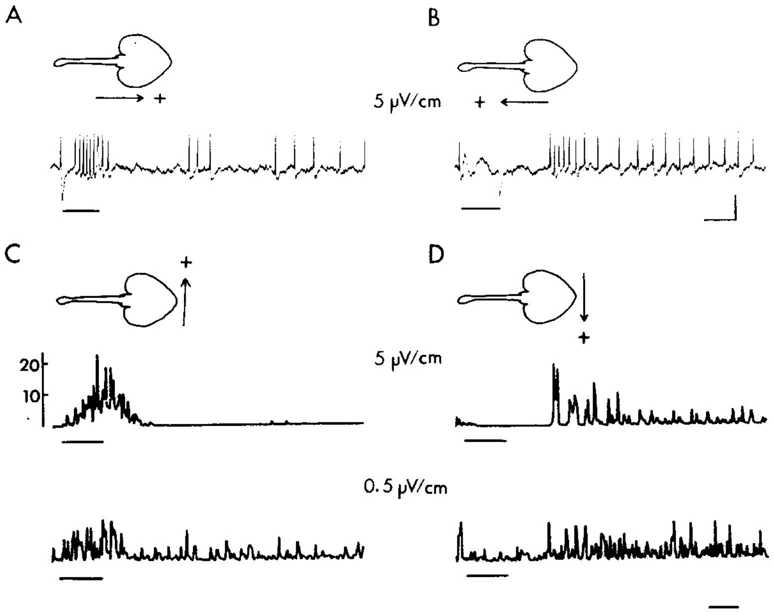

Fig. 2. Dorsal root fiber responses to electric field stimuli. A and B: responses of an ampullary fiber in the dorsal root of the anterior lateral line nerve to a homogeneous electric field of 5 μV/cm oriented parallel to the longitudinal axis of the fish. The electric field orientation and polarity is shown diagrammatically in each case. C and D: post-stimulus–time histograms of responses (20 in each case) of a similar dorsal root fiber to fields of 5 μV/cm (upper) and 0.5 μV/cm (lower). Plotted is the number of impulses versus time. Horizontal bars indicate stimulus period. Calibration: horizontal bars = 50 msec; vertical bar = 200 μV.

stimuli used, but many of the units were sensitive to mechanical stimulation resulting from large dorsal–ventral excursions of the pectoral fins. This sensitivity of the ampullary electroreceptors to crude mechanical stimuli is consistent with earlier reports[13,14].

Fibers of the ventral root of the anterior lateral line nerve were recorded after the overlying dorsal root was transected and reflected. The ongoing activity among ventral root fibers was quite variable and, while some cells had regular discharge rates, most were irregular and had lower spontaneous impulse frequencies (≤ 15 imp./sec) than dorsal root fibers. Some ventral root units had ongoing impulse rates near zero.

Of the 59 ventral root fibers tested, 58 were unresponsive to electric field stimulation at any field orientation or polarity, even when tested with fields as great as 5 mV/cm. Only one unit responded to fields as weak as 20 μV/cm, and we believe its presence can be explained by incomplete transection of the dorsal root. However, 42 of the ventral root units were mechanoreceptive, with bursts of spikes elicited by light mechanical stimulation of receptive fields on the head and pectoral fins (Fig. 3). Most of the remaining 17 cells were not held long enough to allow thorough tests with mechanical stimuli.

The physiological evidence, therefore, confirms anatomical data indicating that the dorsal root comprises fibers exclusively from the electroreceptive ampullae of Lorenzini, whereas the ventral root carries fibers from mechanoreceptors of the anterior lateral line system. Further physiological data from evoked potential measurements are consistent with the hypothesis from anatomical studies that the dorsal nucleus is the primary electroreceptive nucleus of the medulla.

In 5 animals electrode tracks through the medulla at the level of the dorsal and medial nuclei revealed a large, usually biphasic evoked potential response (Fig. 4) following stimulation with weak electric fields (3–50 μV/cm). The latency to onset and profile of this response was dependent on the orientation and polarity of the stimulus field and ranged from 20–30 msec. The maximum amplitude of this evoked wave was

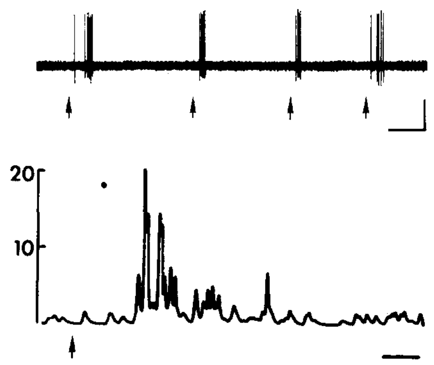

Fig. 3. Ventral root fiber responses to mechanical stimuli. Upper: responses of a ventral root unit to light brush strokes (arrows) to the ipsilateral pectoral fin. Calibration: horizontal bar = 0.5 sec; vertical bar = 200 μV. Lower: PST histogram of 20 responses of a similar unit to water drops (arrows). Plotted is the number of impulses versus time. Calibration: horizontal bars = 50 msec.

found at mid-dorsal nuclear levels, 1 mm below the surface of the medulla. A smaller, longer latency (about 30 msec to onset) response persisted after transection of the ipsilateral dorsal and ventral roots (Fig. 4). This response apparently reflects higher order connections via the contralateral dorsal nucleus, as each dorsal root is known to project only ipsilaterally[3,10].

No significant evoked activity was elicited by electric field stimuli at electrode positions and depths corresponding to the medial nucleus. However, a second averaged evoked potential response with two negative peaks was recorded at depths of 3500–4500 μm below the medullary surface (Fig. 4) at a latency to onset of 22–30 msec; histological localization of recording sites revealed that this activity is associated with the lateral reticular zone of the medulla. This response is likely attributable to

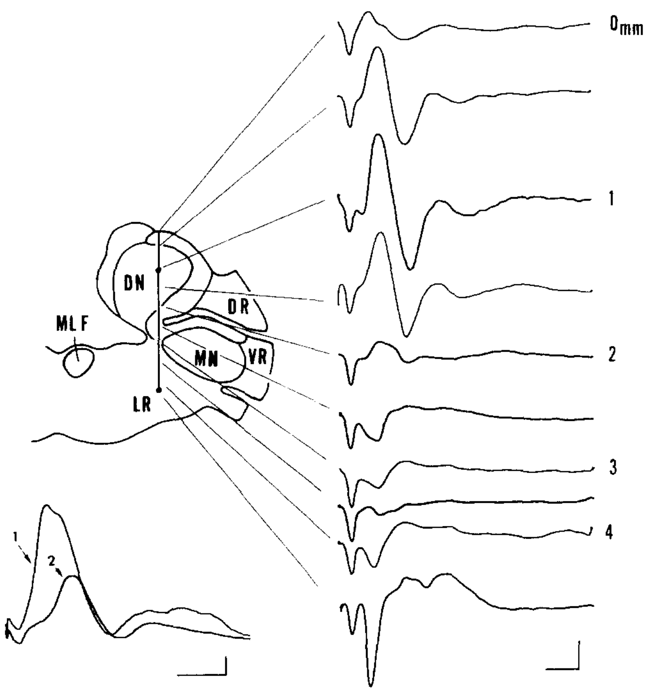

Fig. 4. Evoked potentials from the medulla to electric field stimuli. Responses to a field of 5 μV/cm are shown at right as a function of depth along a single electrode track through the medulla (left) at the level of the lateral line lobes. Filled circles in the electrode track indicate marked recording sites. Each record is an average of 16 trials. Positive potentials are upward deflections and the early negative peak in each trace is a stimulus artefact. While large responses are measured from the dorsal nucleus, no large evoked potentials are measured at electrode positions near or within (in other tracks) the medial nucleus. However, a negative evoked potential is also measured from the lateral reticular zone. At lower left is an evoked response from the dorsal nucleus to a field (50 μV/cm) before (1) and after (2) transection of the dorsal and ventral roots of the ipsilateral anterior lateral line nerve. As each dorsal root is known to project only ipsilaterally, the response after transection must reflect higher order connections via the contralateral dorsal nucleus. Calibration: horizontal bars = 50 msec; vertical bars = 50 μV. Abbreviations: DN, dorsal nucleus of octavolateral area; DR, dorsal root of anterior lateral line nerve; LR, lateral reticular formation; MLF, medial longitudinal fasciculus; MN, medial nucleus of octavolateral area; VR, ventral root of anterior lateral line nerve.

dorsal nucleus efferents terminating in the reticular nucleus as there are no dorsal root fiber projections to this area but the dorsal nucleus efferents are known to pass through the reticular zone in route to the midbrain electroreceptive nucleus[4].

DISCUSSION

Our physiological data confirm an anatomical separation of electroreceptive and mechanoreceptive information in the dorsal and ventral roots, respectively, of the anterior lateral line nerve. The large evoked potential responses to electric field stimuli further support the anatomical evidence that the dorsal nucleus is the primary electroreceptive nucleus in the medulla of elasmobranchs. Platt et al.[19] recorded similar evoked potential responses in the electric ray, Torpedo, following direct electrical stimulation of a branch of the anterior lateral line nerve that innervates primarily ampullary electroreceptors (maxillary nerve)[19]. Although it was not determined if recording sites were in the anterior or posterior lateral line lobe, the peak amplitude of the response was found at a depth of 500–1000 μm below the surface, as in the present study, and a smaller evoked response was also recorded from the contralateral lateral line lobes. Paul and Roberts[18] failed to find such a contralateral evoked response to electrical stimulation of the anterior lateral line nerve in Scyliorhinus, but their recordings were made only from the surface of the contralateral hindbrain.

Evoked potential responses in the lateral line lobes following electrical stimulation of the entire anterior lateral line nerve have been studied in Platyrhinoidis[15] and Scyliorhinus[18]. Electroreceptive fibers of the dorsal root and mechanoreceptive fibers of the ventral root were both stimulated in these experiments, thus the distribution of these sensory modalities in the medulla could not be defined.

Several studies[1,5] have reported responses from the elasmobranch lateral line lobes to weak electric field stimuli in seawater, but the exact recording locations were not indicated. It is noteworthy, however, that medullary responses in triakid and carcharhinid sharks[5] were recorded with fields as weak as 0.015 μV/cm.

While our data indicate that the dorsal octavolateralis nucleus is the primary medullary nucleus receiving ampullary input, they do not exclude the possibility that the dorsal nucleus might also receive mechanoreceptive input. The dorsal root of the lateral line nerve projects directly to a portion of the lateral granule cell mass capping the dorsal nucleus, as well as directly to the dorsal nucleus itself[3,10]. The ventral root of the anterior lateral line nerve also projects to a portion of the lateral granule cell mass, as well as to the medial octavolateralis nucleus[3,10]. Thus the granule cell mass receives ampullary and mechanoreceptive inputs and is known to project back upon both the dorsal and medial octavolateralis nuclei[2]. The lateral granule cell mass also receives primary octavus afferents[17]. It is now known that the primary projections of the octavial, ampullary, and mechanoreceptive inputs are to different portions of the lateral granule cell mass[10,11,17], but it is not known whether this segregation is maintained by the efferent projections of the lateral granule cells back onto the octavolateralis nuclei. Single cell recordings within the octavolateralis nuclei are needed to resolve these questions.

An anterior lateral line nerve divided into dorsal and ventral roots, with the dorsal root projecting to a dorsal octavolateralis nucleus, exists not only in cartilaginous fishes but also characterizes lampreys and chondrostean, crossopterygian, and dipnoan fishes[11,16,17]. On the basis of this pattern we suggest that all of these taxa are electroreceptive and that the dorsal octavolateralis nucleus is the primary electroreceptive medullary nucleus in these fishes as well.

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