

THE RESPONSE OF THE AMPULLAE OF LORENZINI OF ELASMOBRANCHS TO MECHANICAL STIMULATION

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INTRODUCTION

It has generally been assumed since Sand's electrophysiological demonstration of the thermal sensitivity of the ampullae of Lorenzini of elasmobranchs (Sand, 1938) that their function is temperature reception. The simple behavioural experiments of earlier workers (e.g. Parker, 1909; Dotterweich, 1932) had indicated a mechanoreceptive function of some sort, but Sand had been unable to detect a response in the nerves to mechanical stimulation, and Hensel found such a massive stimulus necessary (in dogfish) that a mechanoreceptive function seemed improbable (Hensel, 1956).

However the anatomical arrangement of the ampullae and their tubes, especially in rays (Figs. 1 and 2), cannot be understood if their function be thermoreception, since the jelly in the tubes does not appear to have a specially high thermal conductivity (see p. 422). A further investigation of the function of the ampullae was therefore justified, and a preliminary account of this has appeared (Murray, 1957).

MATERIAL AND METHODS

Various *Raja* species were used, mainly *R. clavata*, but occasionally *R. naevus* or *R. montagui*; the results obtained with each species were similar. A few dogfish (*Scylliorhinus canicula*) were also studied. For convenience, the head of the pithed fish was cut off, but when this was not done, the ventral aorta was ligated to prevent the seepage of blood into the dissection cut. The ampullae of the mandibular capsule were used, because of the ease of the dissection and the length of fine nerve available. In early experiments the nerve, capsule, tubes and overlying skin were dissected out, but later the capsule and tubes were left *in situ* and undisturbed except that the skin over the course of the nerve had to be cut away as far as the base of the capsule. This had to be done because it was essential in testing for mechanical sensitivity to cut the nerve from the more sensitive lateral line organs which forms a mixed nerve trunk with the ampullary fibres (Fig. 2A). The cut can only be made reliably distal to the base of the capsule where the lateralis nerve branch is separate from the ampullary, and lies beside the capsule on its way past from the mandibular lateral line canal. The nerve was lifted up over a pair of Ag/AgCl wire electrodes, and strands were separated off successively with scissors and thinned down until a record was obtained with a suitably small number of

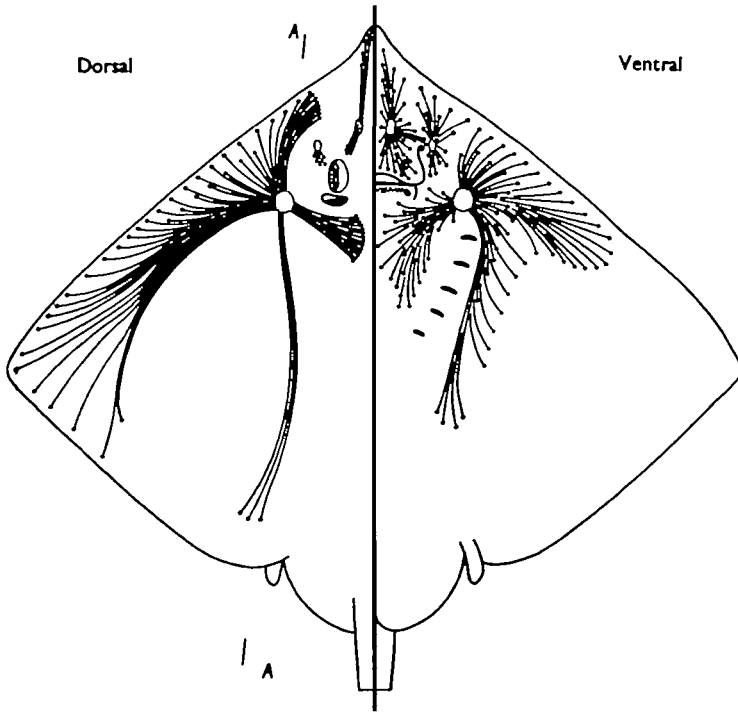


Fig. 1. Diagram showing the extent of the tubes of the ampullae of Lorenzini of an individual *R. clavata*, on the dorsal and ventral surfaces. The tubes are not shown which come from the mandibular capsules just behind the mouth. The other three capsules on each side have tubes opening on both dorsal and ventral surfaces. The section at *A-A* is shown in Fig. 2C.

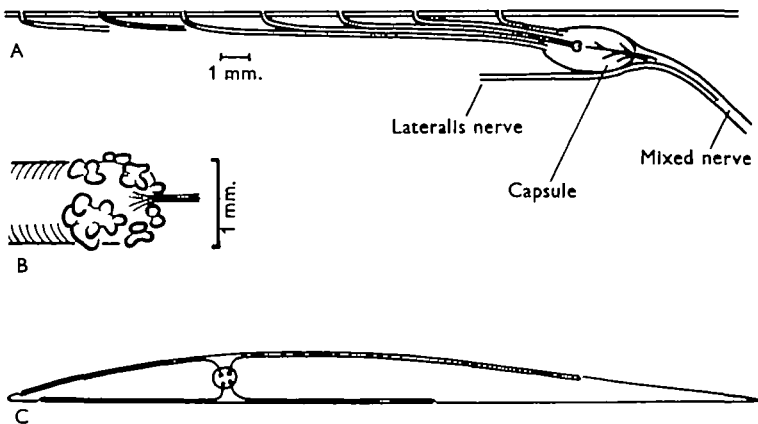


Fig. 2. A. Diagrammatic section of the skin just behind the mouth, showing the mandibular capsule and some of the tubes running from it. The scale refers to a fish of about 50 cm. span. B. An individual ampulla from the hyomandibular capsule of a 50 cm. span *R. clavata*. C. Diagrammatic longitudinal section through the base of the wing (at *A-A* in Fig. 1) showing four of the ampulla tubes and the hyomandibular capsule.

active units. Normally the sensitivity of the preparation would be tested at this stage before the attempt was made finally to isolate a single unit. The process of separation of strands could be repeated along the length of the nerve so that more units could be investigated.

A roughly quantitative assessment of the sensitivity of the preparation was made by using progressively stiffer probes. Initial tests were made with a nylon filament of $30\ \mu$ diameter, which could exert a force of up to the equivalent of 1 mg. If this was inadequate a human hair was used, which exerted a maximal force of the equivalent of 5–50 mg according to the angle at which it was held. Finally, a pointed rod was used. When sensory adaptation was being studied the probe was held in a manipulator.

Nerve impulses were recorded with a conventional a.c. amplifier, C.R.O. and tape-recorder.

RESULTS

The ampullae of Lorenzini in rays are sensitive to very slight mechanical stimulation; when the end-opening of the tube of an ampulla is lightly touched with the nylon filament the resting discharge in the corresponding nerve fibre is briefly speeded-up (Fig. 4A), or a short burst of impulses occurs if there is no resting discharge (Fig. 3). If now another strand of the nerve is used for recording, a different opening is found to be sensitive. In this way, in some preparations, sensitive responses have been obtained from between one-third and one-half of the 20–30 ampullae of the mandibular capsule; this means that on about half the occasions on which the nerve was successfully dissected down to leave only two or three active units a touch-sensitive opening could be found.



Fig. 3. The impulse discharge in a two-fibre preparation showing the brief burst of impulses in the spontaneously silent unit when the corresponding opening is touched with a nylon filament.

The ampullae can easily be overstimulated and their sensitivity reduced or abolished. Touching a previously 'nylon-sensitive' opening with a hair abolishes the response to nylon, at least for a few minutes and often permanently, although the response to a hair may remain. Even 'hair-sensitivity' may be lost if the hair is pushed right into the opening of the tube—a stimulus which produces an impulse discharge at maximal frequency, adapting after several seconds. Because of the ease with which the sensitivity of the organs may be destroyed it is essential that dissection be kept to a minimum. Unfortunately, although the capsule and the tubes can be left intact in their places, the nerve has to be followed up to the capsule so that the lateralis branch may be cut, and this involves some disturbance of the capsule. It is not therefore surprising that many preparations showed reduced sensitivity.

So far, reference has been made only to excitatory effects. Touching the opening results usually in an increase in the frequency of the resting discharge, followed by an inhibitory after-effect when the stimulus is removed (Fig. 4). But often the initial effect is inhibitory, the discharge being slowed down or even stopped during the touch, with a post-inhibitory rebound (Fig. 5). The sensitivity of the two kinds of response is similar. Sometimes an active unit which is speeded up by a touch on one opening may be slowed by touch on the skin alongside or at an adjacent opening.

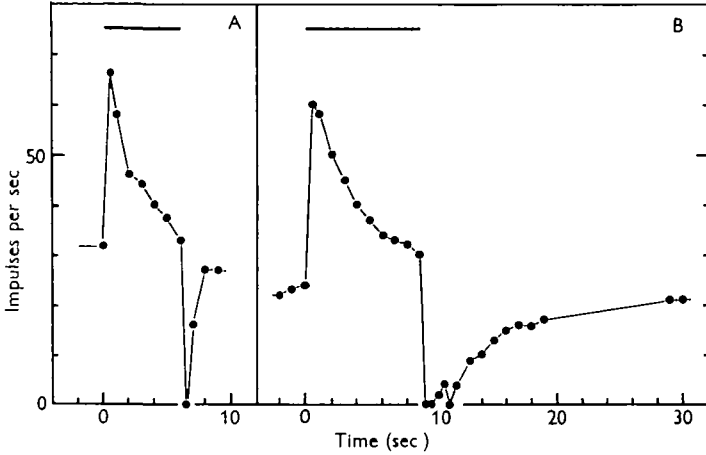


Fig. 4. Discharge frequencies of two preparations in response to touch. The duration of the stimulus is marked by the horizontal line. (Stimulus in A, nylon; in B, rod.)

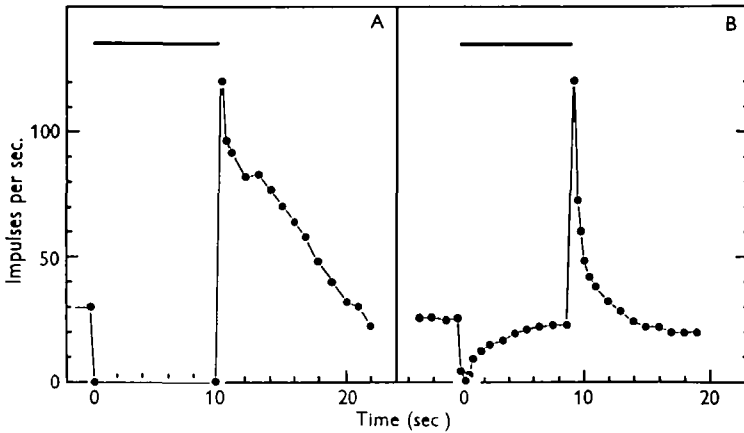


Fig. 5. Discharge frequencies of two preparations in response to touch. The duration of the stimulus is marked by the horizontal line (Stimulus in A, nylon; in B, rod.)

In a series of 15 consecutive preparations (10 of them isolated, 5 *in situ*) 32 nylon-sensitive single units were identified in which the frequency was increased, and 15 in which it was decreased by touching an opening. There were also 19 units

which were speeded by a hair touch, and 4 which were inhibited. The remaining small multi-fibre or single-unit recordings, in which a response to touching an opening could not be elicited, could almost all be excited and inhibited by touching or distorting the region where the tubes emerged from the capsule.

Typical response curves (Fig. 4, 5) show that the process of 'adaptation' is total and relatively rapid, the discharge frequency returning three-quarters of the way back to its initial level in 3-8 sec. However, it is not possible to state that this is a genuine process of adaptation in the sensory nerve ending as it may result in part from mechanical accommodation in the sense organ reducing the effectiveness of the stimulus. For instance, the jelly from the ampullae behaves elastically to sudden pressures, but flows plastically under maintained pressure. In a few preparations (Fig. 6) in which the reduced sensitivity necessitated stronger stimulation such as firm pressure with a blunt rod over the centre of the extent of the tube, the adaptation time was as long as 20-30 sec., which is similar to that found with electrical stimulation (Murray, 1959).

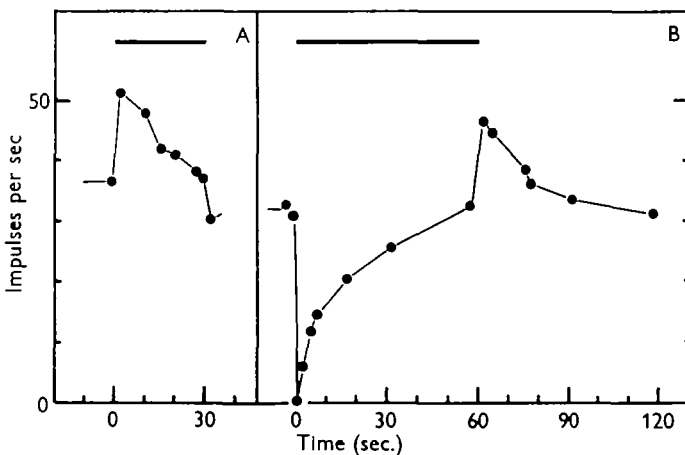


Fig. 6. Discharge frequencies of two preparations in response to maintained firm pressure on the tubes showing the longer adaptation times. The duration of the stimulus is marked by the horizontal line.

Other methods of mechanical stimulation have been observed qualitatively to be effective. If the whole preparation is immersed in sea water, a pipette-jet directed against the opening, strong enough to cause a small visible indentation in the skin, results in a detectable change in impulse frequency. Movement of the skin surrounding the capsule and tubes is also effective: for example, pushing the lower jaw 0.5 mm. upwards (morphologically) or pulling the skin at a point 20 mm. posterior to the organs so that the region of the openings moves 0.2 mm., are both stimuli strong enough to cause complete inhibition of the resting discharge (the opposite movement of course causes increase in frequency). In view of Dotterweich's suggestion that the ampullae are depth receptors, the effect of changing

the depth of the shallow water covering the preparation was studied in a few experiments. No clear results were obtained as although responses did sometimes occur the possibility of movement in the preparation could not be excluded.

Experiments on the mandibular capsules of the dogfish (*Scylliorhinus canicula*) show that they also are mechanically sensitive, but because the openings are hidden beneath the scales, hair or nylon testing is not possible.

However in one preparation, for example, pressure with a light plastic rod near the centre-line of the fish just behind the lower lip was excitatory, and pressure on the skin just beside the front end of the capsule was inhibitory for one particular unit, and the impulse discharge in another was speeded by touch two-thirds of the way to the centre-line along the line of the tubes while pressure just anterior to the fold at the corner of the lip was inhibitory.

In connexion with the possible thermoreceptive function of the ampullae the changes of temperature inside one of the hyomandibular capsules and in muscle at a similar depth in the body of a pithed ray were compared following a sudden change in the temperature of the surrounding water. Thermocouple junctions embedded in hypodermic needles were used. No evidence was obtained of a more rapid change of temperature in the capsule than in the muscle.

DISCUSSION

From their nature, electrophysiological experiments cannot demonstrate the functional importance of a sense organ in the life of an animal. Only if there is a single, clearly most sensitive, modality of stimulation can the function be inferred. This is certainly not so in the case of the ampullae of Lorenzini, where responses sensitive enough to be the basis of a sensory function have been recorded for thermal (Sand, 1938) as well as mechanical stimuli. Moreover, experiments in progress indicate that the ampullae are sensitive to voltage gradients in the water as small as $2 \mu\text{V./cm.}$ (Murray, unpublished; see also the hypothesis of Lissmann, 1958). The real function of the ampullae therefore remains uncertain. The movements of the body of the fish which occur in swimming or feeding will be effective stimuli and so also will contact with the substrate. But the fish has other receptors which between them could cover these functions adequately, such as the muscle stretch receptors (Fessard & Sand, 1937), the dermal pressure receptors (Lowenstein, 1957) and the cutaneous free nerve endings. Like the lateral line organs, which are also sensitive to distortion of the skin resulting from the fish's own movements, but which have in addition a well-circumscribed special function (see Dijkgraaf, 1952), the ampullae must presumably have some distinctive function, and it is reasonable in view of their anatomy and central location to suppose that accurately timed comparisons between what is happening at the ends of the tubes will be important. Evaluation of the changing hydrodynamic pressure distribution over the surface of the aërofoil-like body when swimming or gliding could be such a function.

The following calculation indicates the magnitude of the pressures involved (see Goldstein, 1938). The greatest positive increase in pressure which can occur

at the front of an aerofoil (at the stagnation point a little way back on the under surface) is $\rho V^2/2g$ where ρ = density of the medium, g = the acceleration due to gravity and V = velocity. In a conventional aerofoil, and especially in one as thin as a ray's wing, the decrease in pressure just behind the leading edge may at suitable angles of incidence be as much as four or five times greater than $\rho V^2/2g$. If the angle of incidence changes suddenly, it is reasonable to suppose that changes of pressure could occur of about twice $\rho V^2/2g$, either increase or decrease. If a speed of 100 cm./sec. is assumed, the pressure changes would be approximately 10 g./cm.². Now the area of the opening of the mandibular ampulla tubes is 10^{-4} cm.² and therefore the changes in force applied to one ampulla would be 10^{-3} g. This is the value found to be effective experimentally.

The transducing mechanism, whereby the mechanical stimulus is converted into a change of nerve impulse frequency, also remains obscure. It is clear that stimuli which raise the pressure of the jelly inside the ampulla relative to the general pressure in the capsule are normally excitatory, and that inhibition is caused by stimuli which lower the relative ampulla pressure, as for example skin distortion which stretches the tube, or even touch on an adjacent hole which increases the local capsule pressure. But it may also be that there are specific 'inhibitory' nerve endings, in which an increase of ampulla pressure results in a decrease of frequency. A double innervation is possible anatomically, since about six fibres run from each ampulla.

Sand's inability to detect a response to mechanical stimulation can be attributed to the reduction in sensitivity which occurs when the preparation is disturbed and in particular when it is dissected away from the body. The low sensitivity of Hensel's isolated dogfish preparation (he had to press hard on the capsule wall to get a response, which incidentally was inhibitory) was probably also due to this kind of change. In fact, threshold sensitivity tests are of little value when made on isolated, and so possibly damaged, preparations. But Sand also recorded *in situ* without much dissection, but from the whole nerve, and in this case the mechanical stimulus would have caused as many fibres to be inhibited as were excited and so the response might have been overlooked. Where a stimulus may cause either speeding or slowing of the discharge, it is essential to reduce the complexity of the recording until the response of individual units can be recognized.

SUMMARY

1. The ampullae of Lorenzini are sensitive to weak tactile stimulation applied to the ends of their jelly-filled tubes.
2. Either an increase or a decrease in their resting discharge frequency may be caused, each with an opposite after-effect.
3. 'Adaptation' is total, being three-quarters completed in 3-8 sec. This 'adaptation' probably includes accommodative changes of the tissues.
4. The function of the ampullae is discussed, but no definite conclusion can yet be reached.

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