

LABYRINTH AND EQUILIBRIUM.

I. A COMPARISON OF THE EFFECT OF REMOVAL OF THE OTOLITH ORGANS AND OF THE SEMICIRCULAR CANALS.

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INTRODUCTION.

According to the conceptions of Mach and Breuer we must distinguish two kinds of equilibrial functions in the ear; the one, dynamic through which movements of rotation are perceived, and the other, static by which is produced a definite orientation or sensation of position in relation to the lines of gravitational force. It was suggested that the dynamic function is performed by the sensory structures of the semicircular canals, the static by the otolith organs of the vestibule. Although this view has been widely accepted the literature of the subject is full of contradictions and the experimental evidence is far from satisfactory.

Results of experiments on the labyrinth on which I have been engaged for a long time show that no sharp differentiation exists between the functions of the otolith organ of the utriculus and the ampullæ of the semicircular canals. I have found on the one hand that a labyrinth from which the ampullæ have been removed without injury to the vestibular portions possesses both dynamic and static functions, and that on the other hand a labyrinth from which the otolith organs have been removed without injury to the ampullæ retains both static and dynamic functions.

The experiments reported in this article have been made on dog-fish of the genera *Mustelus* and *Squalus*. The advantages of the dog-fish as an experimental animal have often been pointed out. Among these are the large size of the various parts of the labyrinth, the

clearness with which operations may be performed through the transparent cartilage of the skull, and the fact that bleeding can be almost wholly avoided and that the fish recovers readily from the shock of the operation.

In my experiments it has been necessary to use as indices of results the well known compensatory movements which occur on rotation, the righting reaction, and the general state of equilibrium. The compensatory movements of the eyes and fins of the dogfish, as first described by Loeb,¹ are remarkably constant. I have made especial use of the eye movements which may be described briefly as follows: if a dogfish is held in its normal resting position, belly down, and longitudinal body axis horizontal, the eyes assume a characteristic resting position, symmetrical to the body. This position of the body I shall refer to as normal and the position of the eyes as the primary position. If the animal is rotated to the right around its longitudinal axis, the eyes make complementary movements of such character that they tend to preserve their original position in space; that is, the right eye is elevated and the left eye is depressed. If the rotation is around the transverse axis, head inclined downward, both eyes appear to rotate on their axes by a wheel-like movement so that the anterior pole of each eye is elevated and the posterior pole is depressed. If the animal is rotated backward around its transverse axis both eyes make a compensatory wheel-like forward rotation. If the animal is rotated about an obliquely placed horizontal axis the compensatory movement is a resultant of the effects which would be produced by the rotations around the two axes of reference. These compensatory movements are made during the rotation, but if the abnormal body position is retained, the complementary position of the eyes is also retained. It is seen, of course, that as long as the animal is kept in the abnormal position there is a constant force; namely, the force of gravity, acting upon its body elements in an unaccustomed direction.

If the animal is held in the normal position and the body is then rotated around its dorsoventral axis, both eyes turn by a conjugate movement in the opposite direction; that is, if the head is swung to the right both eyes turn to the left, and if the head is swung to the

¹ Loeb, J., Ueber Geotropismus bei Thieren, *Arch. ges. Physiol.*, 1891, xlix, 175.

left both eyes look to the right. If in these rotations the new position is retained, the new position of the eyes is not retained; they return to their primary position. It is seen at once that in the new position there is no altered relation to the lines of gravitational force.

Removal of the Ampullæ.

The ampullæ of the anterior vertical and the horizontal canals open into the utriculus so near the macula acustica of the latter that most experimenters seem to have had difficulty in destroying the one set of structures without injury to the other. Lee² and Lyon³ each speak of destruction of the ampullæ, but both seem to have relied on section of the nerve branches. By section of the nerve branches, however, they arrived at exactly opposite and fundamentally contradictory results.

After considerable practice I have developed a special technique by which the ampullæ of any or all of the canals may be removed with a minimum of injury and shock to the animal and with results which admit of no uncertainty. A flap of skin is loosened and turned back exposing the appropriate portion of the skull. A thin surface layer of the skull is sliced off with the attachment of some of the neck musculature, thus making visible the parts of the labyrinth through the transparent cranial cartilage. The membranous canal is exposed at a distance not too great from its ampullar enlargement. With a fine pointed pair of curved forceps the membranous canal is grasped as closely as possible to the ampulla and the canal with its ampulla is extracted by a sudden movement, a light quick jerk. Success in this operation depends mainly upon the choice of forceps with the proper curve which bite at the very point, and upon acquiring the knack of removal of the canal by a suitable movement. A too sudden pull will usually break off the canal external to the ampulla, and too slow a movement frequently drags and injures portions of the vestibular structures which it is desired to leave unharmed. When

² Lee, F. S., A study of the sense of equilibrium in fishes, *J. Physiol.*, 1893, xv, 311.

³ Lyon, E. P., A contribution to the comparative physiology of compensatory motions, *Am. J. Physiol.*, 1899-1900, iii, 86.

one has once acquired the knack of this operation the results become absolutely clear. The ampullæ can be extracted one after another with certainty and exactness. In sectioning the nerves one may cut too much or too little; the fiber bundles are scattered, and certainty is impossible. The attempted destruction of the ampullæ *in situ* cannot by any means have the exactness of their complete removal. In many of my earlier experiments I had the ampullæ pasted on a blank leaf of my note book when I wrote down on the same page the results of their extirpation. Under these conditions there can be no doubt as to the correctness of the results. In the summer of 1919, I repeated and extended these experiments at the Marine Biological Laboratory, and on account of the contradictions of previous workers I took occasion to have the experiments witnessed by a number of physiologists and zoologists.

I had found⁴ that removal of the ampullæ of the four vertical canals had little or no effect on the compensatory eye movements resulting from the rotation around the longitudinal and transverse axes. In order, however, that there could remain no possible functioning of the semicircular canals I have in a long series of animals removed all six ampullæ with uniform results.

A dogfish from which all six ampullæ have been removed shows definitely the following reactions. (1) Compensatory movements of the eyes and fins occur on rotation around a longitudinal axis; *e.g.*, on rotation to the right, the right eye goes up and the left eye goes down. This position of the eyes is retained as long as the abnormal body position is continued. (2) Compensatory movements of eyes and fins occur on rotation around the transverse axis; *e.g.*, when the animal is tilted head downward the eyes make the characteristic wheel-like backward rotation. (3) Compensation is absent on rotation around the dorsoventral axis. (4) The animal swims in a manner differing but little from the normal. (5) The righting reaction takes place promptly and vigorously; if the animal is placed belly up in water it turns over at once.

As a sample experiment I quote *verbatim* the following from my notes.

⁴ Maxwell, S. S., Experiments on the functions of the internal ear, *Univ. California Pub. in Physiol.*, 1910-15, iv, 1.

"July 14, 1919. Dogfish 5. 10.00 a.m. All six ampullæ removed. Compensatory movements prompt on rotation around longitudinal and transverse axes; none on rotation in horizontal plane. Animal rights itself perfectly in water. Eyelids sewed together to exclude retinal stimuli, and animal put into deep tank; righting perfect.

2.00 p.m. Animal rather weak but rights itself promptly when turned over in water; swims rather wobbly; turned completely over once when excited by other dogfish; I have seen a normal dogfish do this under similar circumstances.

July 15, 9.00 a.m. Animal very weak; rests on bottom of tank in normal position. Rights itself but may swim one or two turns belly up before getting over. Opened stitches in eyelids. No compensatory movements of eyes.

July 16, 9.30 a.m. Animal moribund. Killed for autopsy. Considerable blood clot in each vestibule."

The above experiment shows a possible source of the confusion in the reports of previous investigators. Had I assumed that on account of shock effects observations made on the day of operation would be unreliable, and had I waited until the following day to make my observations, it would have appeared that loss of the ampullæ abolishes compensatory movements, which is manifestly not true. When immediately following the destruction of an organ a function is clearly retained it is indisputable proof that at least that organ is not the only one which can perform the function. Observations made on July 15, on Dogfish 5, might have favored the statement that destruction of the ampullæ of the semicircular canals abolishes compensatory movements of the eyes, but the observations of July 14 clearly show such a conclusion to be wrong.

In the attempts to determine the rôle of the various sense organs in the geotropic reactions of the dogfish, it has long been recognized that retinal stimuli play a part. Lyon³ and Parker⁵ excluded visual stimuli by section of the optic nerve. I have accomplished the same result by the less radical operation of sewing the eyelids together, when equilibrium and the righting reactions were under consideration, and by placing a black, opaque disk on the cornea over the region of the pupil when eye movements were to be studied. Other methods of blinding were also used. I can affirm with complete assurance

⁵ Parker, G. H., Influence of the eyes, ears, and other allied sense organs on the movements of the dogfish, *Mustelus canis* (Mitchill), *Bull. Bureau of Fisheries*, 1909, xxix, 43.

that the compensatory motions described in the case of animals from which all the ampullæ have been removed occur also when activity of the retina has been excluded.

It must also be noted that the dogfish, like most animals which rest on the bottom and are not merely suspended in the water, manifests very strong contact reactions. A vigorous specimen which has been blinded and which has had as far as possible all the end organs of the eighth nerve destroyed will almost always be found belly down when at rest. Such a fish may swim indifferently back or belly up, but when it comes to rest the position is a fair index of the general state of the animal. When an investigator affirms that his specimen came to rest indifferently in any position, he has given good incidental evidence as to the animal's physical condition. In stating that a dogfish deprived of its six ampullæ makes normal righting reactions I have not been unmindful of these facts, but have taken care to exclude the possibility of contact stimuli.

Although it can be proved that after the loss of all the ampullæ, with exclusion at the same time of retinal and contact stimuli, the dogfish makes normal compensatory movements of the eyes and fins to rotations in all vertical planes, it is necessary to note that there are some differences between this and a normal animal.

The following seem to be fairly constant results. (1) The compensatory movements of the eyes, though prompt, are noticeably slower than in the uninjured animal. Compensatory movements due to visual stimuli alone are so much slower, requiring several seconds or even minutes, that no difficulty is experienced in distinguishing these from reflexes of labyrinthine origin. (2) If seized while in the water the animal strongly resists the attempt to turn it back downward. One feels, however, that the resistance is neither as prompt nor as strong as in a normal animal. (3) In swimming there is more or less evident a slight tendency to sway from side to side around the longitudinal axis, like a boat insufficiently ballasted.

These three conditions are less noticeable in vigorous specimens; they become very marked in weakened individuals. They can perhaps all be accounted for by a lowering of muscle tonus. It is important to note that, as I shall show later, precisely the same complex of conditions can be brought about through a totally different operation.

Removal of the Otoliths.

In the dogfish the otoliths are of soft, friable, calcareous material. In the sacculus there is a large otolith spread over the main macula acustica and a smaller mass on the lagena. These are so situated that their removal can be accomplished with little injury and the operation is relatively easy. For the otolith of the utriculus the case is very different. This otolith lies in the recessus utriculi so close to the openings of the ampullæ of the anterior vertical and the horizontal canals that it requires some skill and much practice to remove it without injury to the ampullæ. If, however, after opening the vestibule by removing a portion of the cartilaginous roof, the utricular wall is slit open with a very sharp microdissection knife, the otolith material may be washed out by the careful use of a fine pointed pipette. In a similar way the saccule may be slit open and its otoliths removed. No operation was considered successful unless it was found at postmortem examination that no otolith material remained. For reasons to be stated in another paper it was considered important not only to avoid injury of the ampullæ but also to reduce the injury of the utriculus to a minimum.

After removal of all the otoliths from both ears in successful cases the following results are seen. (1) Compensatory movements of the eyes are made in the regular way to rotations about all three body axes, longitudinal, transverse, and dorsoventral. If the animal is rotated around a longitudinal or transverse axis and held in the abnormal position the compensatory position of the eyes is retained, but when the rotation is around the dorsoventral axis the eyes make the compensatory movement and then return to the primary position. These movements appear to differ from those in the normal animal only in being slightly slower. (2) The animal swims in normal orientation and maintains its equilibrium in the water, but its swimming, like that of the fish without ampullæ, is likely to be accompanied by a rocking movement; this rocking or swaying is less apparent in vigorous specimens. (3) If turned belly up in the water, the fish rights itself promptly; in doing so, however, it sometimes overcompensates and turns almost or completely over.

It will be seen that these results are strikingly similar to those

produced by loss of the ampullæ. It is especially noticeable that there is the same apparent slight slowing of the reactions and the same indication of lowering of muscle tonus in general. In one important respect, however, the result of this operation differs from that of removal of the ampullæ; namely, the compensatory movement to rotation about the dorsoventral axis is retained. It should be stated that in these observations due care was taken to eliminate retinal and contact reactions.

Parker⁵ removed the saccular otolith by way of an opening in the roof of the mouth. He found that the loss of this otolith alone produced no noticeable effect on the equilibrium or the righting reactions of the dogfish, nor did there appear to be any loss of tonus. I have removed this otolith many times by the method I have described above. Its loss does not alter or weaken any of the compensatory movements; it does not disturb the equilibrium or the righting reaction, nor is the muscle tonus affected to any noticeable degree.

Removal of Both Ampullæ and Otoliths.

I may say at once that in accordance with the findings stated in the last section the presence or absence of the large otolith of the saccule is without influence on the equilibrium reactions and it may be disregarded. If, however, the utricular otolith has been successfully removed and the condition described in a previous section has been attained, namely the retention of compensatory movements to rotations in all planes, the righting reaction, and the maintenance of equilibrium, the consequent removal of the six ampullæ produces at once a profound alteration. The condition of a dogfish deprived of the utricular otolith and the six ampullæ may be stated in the following way. (1) No compensatory movements are made on rotation around any axis whatever. This statement may be modified by saying that in some cases a slow and slight tendency to compensation, requiring many seconds or even minutes for its completion, may be seen. No one familiar with the reactions of the animal would ever confuse this with a labyrinthine reflex. (2) The animal shows no tendency to maintain bodily equilibrium; it swims indifferently back or belly upward. A weak specimen may also come to rest on its

side or back, but a vigorous specimen usually rights itself on the bottom of the tank. In other words the geotropic reactions of the animal are definitely and completely lost; the stereotropic reaction is retained.

CONCLUSION.

The results of my experiments show that the assumption of a sharp differentiation of function between the otolith-bearing, vestibular portions of the labyrinth and the semicircular canals is not justified by the facts. Between the effects of extirpation of the one and of the other set of structures there is more resemblance than contrast. They certainly reenforce each other, for the reactions produced by either one alone are always slower and less vigorous than when both sets of organs are intact. It would not, however, be safe to affirm that the functions are identical. In one respect a difference is apparent; namely, in the response to rotation in a horizontal plane. If the ampullæ are uninjured, compensatory movements occur when the animal is rotated around its dorsoventral axis. I have never seen this reaction in the absence of the ampullæ of the horizontal canals.

SUMMARY.

1. A dogfish from which all six ampullæ have been removed maintains its equilibrium; the righting reactions occur promptly; compensatory movements of the eyes occur in response to rotations in all planes except the horizontal; the compensatory position of the eyes is retained if the animal is held in an abnormal position. Both the static and dynamic functions of equilibrium continue, therefore, after complete removal of all the semicircular canals and all the ampullæ.

2. After complete removal of the otoliths from the vestibules without injury to the ampullæ the animal maintains its equilibrium in the water, rights itself promptly, and makes compensatory motions to rotations in all planes. If held in an abnormal position the compensatory position of the eyes is maintained. Both static and dynamic functions of equilibrium continue.

3. Destruction of both the semicircular canals and the otolith organs completely abolishes all compensatory movements and equilibrium reactions of labyrinthine origin.

4. It is pointed out that these observations do not justify the theory of Mach and Breuer that the ampullæ and semicircular canals are the organs for the dynamic functions of equilibrium, and that the otoliths are the organs for the static functions of equilibrium.

5. The new experiments recorded in this paper show that the ampullæ alone (without the otoliths) suffice for all the dynamic and all the static functions of equilibrium of the ear; and that the otolith organs (without the ampullæ) suffice for all the static and for all the dynamic functions of equilibrium of the ear with the exception of the response to a rotation of the animal in a horizontal plane.