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CUPULA MOTION IN THE SEMICIRCULAR CANAL OF THE SKATE, *RAJA ERINACEA*

An Experimental Investigation

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Abstract. An upper bound on the magnitude of semicircular canal cupula motion was experimentally determined in the isolated labyrinth of the skate, *Raja erinacea*. To visualize the cupula, a glass pipette was pushed through the ampullary wall, and local regions of the cupula were stained by slow infusion of small amounts of Alcian Blue dye. Carloric stimuli which produced large changes in single unit activity in the ampullary nerve and which often recruited several larger, previously silent units were found to produce no detectable cupula motion as seen through the ampulla wall. However when the cupula was first grossly displaced, motion was thereafter observed in response to identical caloric stimuli. Analysis of afferent responses indicates that the normal range of cupula motion in the skate is below the optical resolution of the method, conservatively estimated as 3–5 micrometers.

The question of what constitutes the naturally occurring dynamic range and mechanical mode of cupula displacement is one of the oldest problems in vestibular physiology, one that cannot be satisfactorily resolved on the basis of anatomical studies. Over the years, investigators have therefore attempted to study cupula motion *in vivo,* using a variety of staining and contrast-enhancing techniques to improve the visibility of the normally transparent cupula.

In early experimental work (Steinhausen, 1927, 1931, 1933; Dohlman, 1935; also see Trincker, 1962), the cupula was described as bending so that its upper surface moved along the vault of the ampulla in a manner of a swinging door. However, more recently, they hypothesis that "swinging door" motions typify the normal cupula bending mode has been questioned (Vilstrup, 1950; Dohlman, 1971; Money, 1971). Hillman (1972, 1974) and his collaborators (Llinas & Hillman, 1973; McLaren & Hillman, 1976) have reported that the cupula in the bullfrog appears to have a circumferential attachment. Several authors (deVries, 1956; Dohlman, 1969, 1971; Hartmann & Klinke, 1975) have also suggested that normal cupula movements must be considerably smaller than anticipated by early workers. A theoretical study (Oman & Young, 1972) indicated that cupula motion in the human semicircular canal should be on the order of a few microns in response to maximal self-induced sinusoidal head motions. Nevertheless, in some recent studies on isolated semicircular canal preparations (Harada, 1972; Schmid et al., 1973; Taglietti et al., 1973) it has been tacitly assumed that the cupula normally makes "swinging door" motions well in excess of several microns, a view that has also been perpetuated in many contemporary texts on vestibular function and anatomy.

In this paper, we describe our efforts to determine whether one can see motion of the stained cupula by looking through the wall of the ampulla while subjecting the canal to moderately large caloric stimuli. Resolution that can be achieved with the light microscope is approximately 0.5 μm, and when one observes the cupula through the wall of the ampulla, optical resolution is considerably de-

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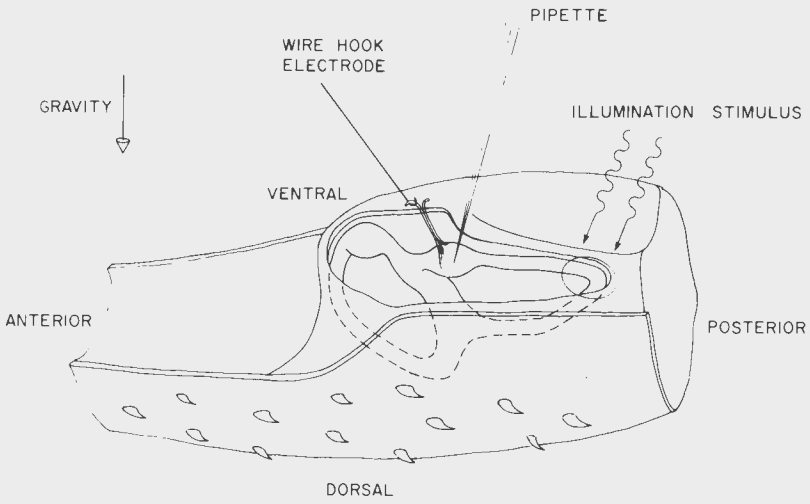

Fig. 1. Schematic diagram of isolated (*right*) labyrinth of the skate, showing anterior and horizontal canals and ampullae exposed on lateral side. Tip of pipette containing dye solution is in the horizontal ampulla. Microscope illuminator produces a spot of light on part of the horizontal canal duct, resulting in an excitatory caloric stimulus.

graded. Therefore, we anticipated that under favorable conditions with large stimuli, movement of the cupula would be difficult to visualize optically through the ampulla wall, while threshold motions should be well beyond the range of light microscopy. For our study of cupula motion, we developed an isolated preparation of the labyrinth of the common skate *Raja erinacea*, and employed caloric (thermal) stimulation, rather than a rotational stimulus because of the technical difficulties anticipated in reliably observing very small cupula motions in a rotating preparation. Caloric stimulation has been used by others in semicircular canal preparations (Steinhausen, 1931; Dohlman, 1938; O'Leary, 1970; Lifschitz, 1973).

In a series of preliminary experiments, we determined that the caloric stimulation technique provided a much smaller, less traumatic stimulus than that resulting when the canal was cut, cannulated, and the cupula hydraulically driven[1] after the method introduced by Steinhausen (1933). In the skate, even in favorable cases the sensitivity and spontaneous activity usually deteriorated soon after cannulation. On the other hand, when the closed membranous labyrinth was stimulated calorically at fixed intervals using the method described below, neural responses remained stable in character for many hours.

METHOD

Male and female skates of approximately 12-inch span were trawled from Massachusetts Bay by commercial fishermen, and kept in sea water tanks at 14°C for periods up to several weeks prior to use.

At surgery, the spinal cord was severed above the first vertebra, the animal was pithed, and the chondocranium was opened dorsally along the midline. The cranial nerves were gently cut, and the brain removed. The floor of the cranium was then divided along the midline of the foramen magnum, and the cranium was sectioned laterally through the orbit. The eye, extraocular muscles, and other tissue were removed on the lateral side. The portions

[1] Injecting fluid caused the stained cupula to deflect visibly in the fashion of a swinging door, apparently free of attachment to the walls of the ampulla. In favorable cases, threshold response as judged by visual and auditory criteria of change in nerve activity occurred for fluid injections of 0.01 to 0.1 μl, corresponding to a deflection at the center of the cupula of 4–40 μm. Infused dye was often maintained entirely on one side of the cupula. Larger hydraulic stimuli could then cause deflections so large that dye was seen to leak across the top of the cupula. In several preparations, the cupula was seen to be torn or detached after the canal duct was cut, cannulated, and dye infused. This is understandable, since theoretical calculations (Oman & Young, 1972) predict that the application of even 0.0001 cm of water pressure across the ampulla would subject the human cupula to an abnormally large steady-state pressure.

Table I. *Composition of solutions (mM/l)*

Composition	MBL standard skate Ringer[a]	Perilymph simulation	Endolymph simulation
NaCl	250	250	200
KCl	4	4	82
$CaCl_26H_2O$	5	3.3	3.8
$MgCl_26H_2O$	2	1.2	0.5
Urea	330	330	330

[a] Buffered to pH 7.4 with Na_2HPO_4.

of the cranium containing the inner ear capsules were excised by severing the hyomandibular cartilage and the vertebral joint and then each was placed in a bath of Ringer's solution. The tissue was kept continuously immersed throughout the experiment in order to avoid subjecting the normally nearly neutrally buoyant membranous labyrinth to unusual forces which might otherwise occur due to drainage of fluid from the perilymphatic space. Sudden changes in temperature were avoided. The preparation was maintained at +14°C or −2°C by means of an icewater bath surrounding the dish.

A lateral window into the cartilaginous labyrinth was cut so that portions of the horizontal and anterior semicircular canals, their ampullae, and associated nerves were exposed. Bundles of ampullary nerves were dissected. Single and multi-unit activity were readily recorded from nerve twigs raised from the bath on a platinum–iridium wire hook electrode, using a high impedance (1 MΩ), high gain preamplifier (Tektronix 5A22N). Amplifier output was bandpass filtered from 0.15 to 10 kHz. A spike amplitude window analyzer (Mentor N750) was used to detect amplitude. The number of spikes detected in this way was counted over 2-sec intervals by an appropriate histogram program running in a (Digital Equipment Corporation Lab 8/E) computer. Amplifier output and voice commentary were also recorded on a multi-channel wideband F.M. tape recorder for further analysis of single unit response. Single units were isolated during repeated tape playbacks by inspection of a storage oscilloscope display, sorted using the window analyzer, and counted by the computer system.

The ear was pinned in place in the dish so that the canal and under stimulation was approximately in an earth-vertical plane with the ampulla uppermost, as shown in Fig. 1. The preparation was stimulated by focusing a small spot of light (1–2 mm ∅) from a microscope illuminator directly on a region of the canal duct. The direction and magnitude of the caloric stimulus could be controlled by positioning the spot of light on either the utricular or the canal side of the ampulla, by varying the intensity or size of the light spot, the duration of the illumination, or by altering the orientation of the canal under stimulation. When, in a given preparation, these variables were kept constant, a highly repeatable stimulus resulted, as judged by the constancy of the neural responses produced.

When initial preparations were complete, and spontaneous activity and sensitivity to caloric stimulation had been demonstrated, a sharply bevelled glass pipette (10–20 μm tip diameter), filled with a solution (see Table I) chosen to match elasmobranch endolymph (Fange et al., 1972; Peterson, 1976), containing 0.5–1.0% Alcian Blue dye, was pushed against the wall of the ampulla, dimpling it. The ampulla was then gently grasped with forceps, and pulled onto the pipette. Local regions of the cupula were visualized by gently infusing very small amounts of dye solution. Stained portions of the cupula were observed with the aid of a calibrated reticule mounted in one eyepiece of a dissection microscope. Diffuse illumination of the entire preparation was provided by a fiber optic illuminator. Caloric stimulation resulting from the fiber optic illuminator was found to be negligible; it was left on for the duration of the experiment.

Response was studied in 59 isolated labyrinth preparations from 54 animals. Cupula motion was produced using a cannulation

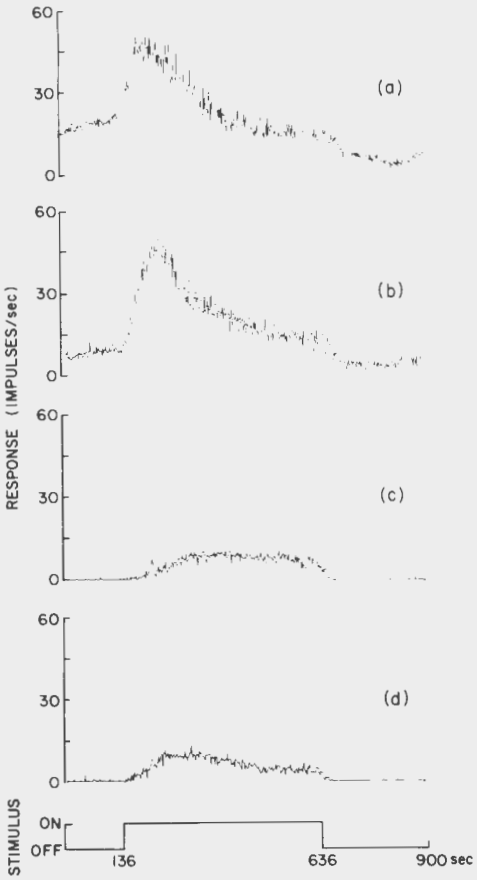

Fig. 2. Simultaneously recording responses of 4 single units (*a*, *b*, *c*, *d*) to the same illumination stimulus (indicated in bottom trace). *Ordinate:* Average frequency of impulses over 2-sec intervals. The slight upward drift in activity of units *a* and *b* apparent prior to stimulus onset is due to residual response resulting from a 300-sec caloric stimulus administered 600 sec earlier.

method in 30 preparations, and by using the caloric/puncture technique in 13 experiments; the results were then compared.

RESULTS

The longevity of the excised preparation was strongly affected by the ionic composition of the Ringer's solution in which the ear was immersed: When MBL Standard Skate Ringer (Table I; Cavanaugh, 1964) was used, spontaneous afferent activity in the nerve usually disappeared after 2–3 hours. However, when calcium and magnesium concentrations were adjusted (as shown in Table I) to more closely match their values in elasmobranch perilymph (Fange et al., 1972; Peterson, 1976), spontaneous and evoked activity often persisted for over 24 hours. The pH of the simulated perilymph was 5.5; buffering to pH 7.4 caused no obvious changes in longevity.

With the extracellular recording technique used, peak-to-peak amplitudes of spontaneously active units ranged up to 500–600 μV. These units varied in their regularity and in their mean spontaneous rate, which typically was 10–20 spikes/second. During caloric and direct hydraulic stimulation of the canal, as response increased, previously silent units were recruited, usually in sequence of increasing spike height. The amplitude of these units ranged up to 1.5 mV.

Typical single unit responses to a sustained (500 sec) caloric stimulus are shown in Fig. 2. Units *a* and *b* were spontaneously active, and of nearly equal amplitude. Units *c* and *d* were initially nearly silent, and had spike heights 80 and 110% larger than Unit A, respectively. The number of nerve spikes observed during successive 2-sec intervals is plotted against time for each unit. Stimulus level (on/off) is indicated in the lowermost trace. Response of spontaneously active units such as *a* and *b* to caloric stimulation was usually apparent within several seconds, increased to a peak 60–100 sec after stimulus onset, and then gradually declined to a rate which in most cases remained above the previous spontaneous level. After the illuminator was switched off, the response declined rapidly. If the duration of the stimulus was sufficiently long enough that the unit response peaked and then declined, the frequency of the response undershot the prestimulus spontaneous rate when the stimulus was turned off. Return of activity to the prestimulus level usually required several hundred seconds. Silent units exhibited higher thresholds to stimulation, and were usually recruited in order of increasing nerve spike amplitude.

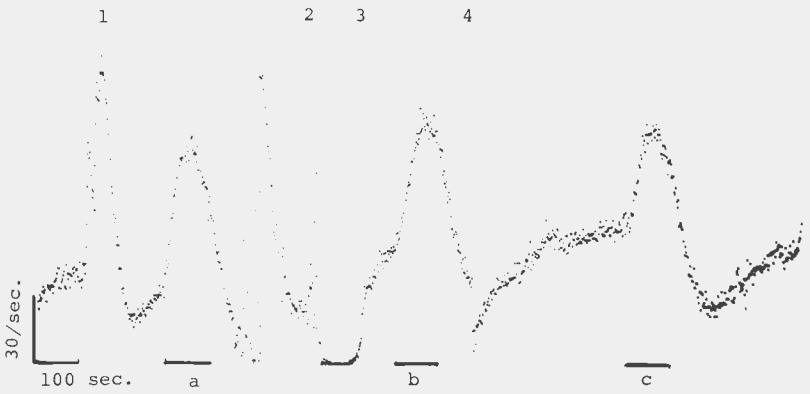

Fig. 3. Frequency of multi-unit response showing effect of puncture of ampulla with dye-filled pipette on successive caloric responses. Average frequency of impulses over 2-sec intervals is plotted vs. time. Stimulus durations, 100 sec, as indicated by bars *a*, *b*, and *c* on abscissa. Run duration: 1800 sec. Pipette pressed on ampulla produced large transients at 1, and again after first caloric stimulus (*a*). Pipette penetrated ampulla at 2, producing inhibitory stimulation. Moving pipette back at 3 restored spontaneous level. Pipette was bumped accidentally at 4. Caloric stimulation showed that peak rate and time course of responses were essentially unaltered by penetration, although spontaneous rate, dependent on pipette position, was somewhat higher after penetration than before.

Horizontal canal units always became excited when the light stimulus was positioned on the canal side of the ampulla, supporting the hypothesis that utriculopetal endolymph movement was thereby produced. When the stimulus was placed on the utricular side of the ampulla, inhibitory responses always resulted. Units from the anterior and posterior canals showed directional sensitivity opposite to that just described. When the canal duct was positioned in an earth-horizontal plane, all units tested were found to be unresponsive to caloric stimuli. Increasing the intensity of the illuminator output increased the magnitude of the response.

Most of the present experiments were conducted using excitatory stimuli, since the low level of activity of spontaneous units in the skate limited the inhibitory response expected to a small dynamic range, and because silent units could be recruited only with excitatory stimuli. The total activity of several simultaneously recorded units was taken as a convenient measure of overall afferent response, and is termed "multi-unit activity" in Figs. 3 and 4. The long-term stability of the closed preparation under caloric stimulation provided the basis for an important control in the present study of cupula motion: We were able to observe regions of stained cupulae under conditions in which the sensitivity of ampullary nerve afferents to stimulation was observed to be essentially unchanged over long periods by the puncture and staining procedures. Successive identical caloric stimuli, presented at equal intervals, usually produced very similar responses.

Comparison of afferent responses to identical caloric stimuli before and after penetration of the ampulla (Fig. 3, *a* and *b*) as well as before and after dye injection (Fig. 4, *c* and *d*) permitted the assessment of the effect of these procedures on the sensitivity of the system. Irreversible changes in response magnitude or dynamics would presumably indicate a significant change in the state of the preparation. Occasionally, upon penetration, a change in the spontaneous level of activity was seen (Fig. 3, event 3) but this could usually be reversed by adjusting the position of the pipette. To prevent accidental overstimulation during dye infusion, nerve response was monitored

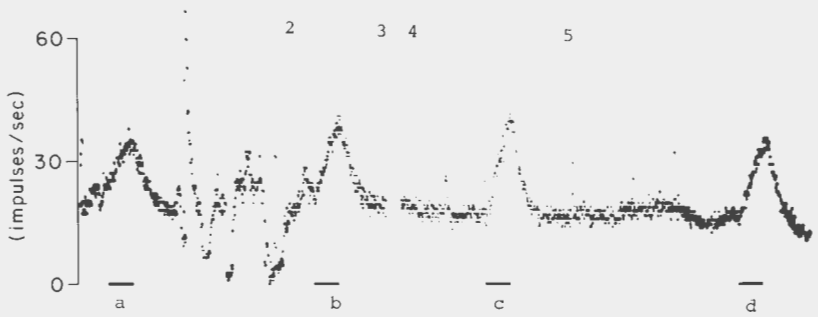

Fig. 4. Frequency of multi-unit response showing effect of ampulla puncture and dye infusion on successive caloric responses. Average frequency of impulses over 2-sec intervals is plotted vs. time. Stimulus durations, 100 sec, as indicated by black bars and on abscissa. Pipette pushed on ampulla at 1 after first caloric stimulus (*a*), and was observed within ampulla at 2. Dye infusion began at 3, producing occasional weak transients in activity. Between 3 and 4 is a data gap of 55 sec while a new computer run was initiated. Dye injection continued at 5, after third caloric stimulus (*c*). No motion of the cupula was seen. Total run duration, 3 100 sec.

continuously through a loudspeaker. The pipette penetration and dye infusion procedures were judged to be successful only if the magnitude and time course of evoked multi-unit response was essentially unchanged by the puncture and injection. With practice, these procedures could be readily accomplished successfully.

After staining, with the pipette still in the ampulla, the cupula was observed in profile monocularly through the eyepiece containing the reticle. The region of the cupula most easily visualized in the horizontal canal was where the cupula meets the vault of the ampulla. In several preparations, it was also possible to look for motion of points near the center of the cupula and near its base. Specific particles of dye were observed at 40× magnification during repeated excitatory caloric stimulation lasting 100 sec (Fig. 4). These stimuli were large enough to double or triple the rate of multi-unit activity in the nerve, and in most instances to recruit several larger units. No detectable motion of any stained region was observed in any of the successful preparations. In several instances the illuminator diaphragm was opened to produce larger and larger stimuli on each successful trial. Even when the diaphragm was completely open, and single unit responses as large as 40 spikes/second resulted, no motion was observed. Optical resolution through the ampulla wall was conservatively estimated at 3–5 μm.

On two occasions when no motion was seen, additional dye was therefore deliberately rapidly injected. Very large afferent responses resulted, and in one case the cupula was seen to move abruptly. Caloric stimuli identical to those which earlier produced no visible displacement now caused the cupula to deflect up to 20 μm in the region of the roof of the ampulla, suggesting that a reduction in the effective mechanical stiffness of the cupula had somehow occurred (see Discussion).

Location of the stained regions relative to the entire cupula was later verified by injection of additional dye. If more dye was infused slowly, the face of the cupula nearest to the pipette gradually became outlined. If injection was very rapid, as described above, the dye solution was often seen to pass between the ampulla and the cupula. Only then

did the Alcian Blue dye appear to penetrate the entire cupula, and to outline its canalicular structure. However, when the cupula was internally stained in this way, unit sensitivity to caloric stimulation was observed to diminish rapidly. After some time, the dye in the ampulla was no longer uniform in distribution, but rather revealed a jellylike granular aggregate of stained material previously not visible in the ampulla.

Cupulae, as seen after Alcian Blue staining, were typically 1.2 mm high (from crista to ampulla vault), 1.4 mm long (in the plane of the crista) and completely covered at least the top of the crista. The cupula was more than twice as wide in the regions of the plana semilunata as in regions above the center of the crista. Cupulae were observed both in situ and after removal from the ampulla. When viewed from above, the outline of the two visible faces of the cupula had the appearance of a pair of parentheses back to back:) (.

DISCUSSION

The results of preliminary experiments in which the canal duct was cut and a cannula inserted were often characterized by a rapidly deteriorating response. In contrast, in experiments where the membranous labyrinth remained closed, spontaneous activity and evoked responses appeared to be much more stable, and to deteriorate rapidly only when abnormally large cupula displacements were created. Thus there seemed to be an important advantage to experimental techniques in which the membranous labyrinth remained closed. Presumably, the canal duct then continued to serve its normal function of equilibrating pressure differences across the ampulla. If a large cut is made in the canal duct, this protective mechanism can no longer operate. Excessive cupula motion may therefore result from cutting the canal or from cannulation. Using the puncture technique, the hole created (typically about 20 μm in diameter) was immediately blocked by the presence of the tapered pipette. Thereafter, even if the pipette was withdrawn, no significant change in the characteristic response to caloric stimulation was observed, probably due to the relatively small size of the opening.

The chemical composition of the artificial perilymph in which the preparation was immersed throughout these experiments appeared important in maintaining its physiological state. Reduction in calcium concentration from 5.0 mM to 3.3 mM, and in magnesium concentration from 2.3 mM to 1.2 mM was found to have a marked effect on the stability of the preparation with respect to maintenance of spontaneous and evoked activity. Concentration changes in the bathing fluid probably reach the nerve fibers and the hair cell/neuron synapses, but due to tight junctions at the lumenal surface, may not reach the apical ends of the receptor cells. The basis for the observed effects of small changes in calcium and magnesium ion concentrations in this system is unknown. However, these ions are critical for the generation of receptor, synaptic, and action potentials in other systems, and are known to affect responses in hair cell systems as well (Russell, 1971; Sand, 1975).

The mechanism of caloric stimulation in man has been well described from a physical point of view (Barany, 1906; Schmaltz, 1932; Steer, 1967; Young, 1972; Demers, 1975). The density of human endolymph is known to decrease with increasing temperature. If the semicircular canal is not at a uniform temperature, in the presence of gravity, the resulting density differences around the canal can produce a convective torque on the ring of endolymph, and hence also on the cupula, physically equivalent to an angular acceleration stimulus. The dynamic response of afferent units observed in this preparation was consistent with the postulated mechanisms of caloric response in man. The observed response of ampullary afferents therefore probably reflects primarily the effect of heat transfer in the tissue and bath operating upon the

torsion pendulum dynamics of the cupula–endolymph system.

The response characteristics of afferent nerve fibers from the semicircular canals have been studied in a number of other species in recent years with regard to both spontaneous and evoked activity (Goldberg & Fernandez, 1971; Precht, Llinas & Clarke, 1971; Correia & Landolt, 1973; O'Leary et al., 1974; Blanks et al., 1975). The time course of response to caloric stimuli observed in afferent units in the present experiment appeared qualitatively similar to that reported in other preparations to steps of constant angular acceleration if a short lag in response due to the (nonlinear) dynamics of the heat transfer processes were taken into account. As in the other studies, units observed varied significantly in terms of the rate and regularity of their spontaneous activity, the magnitude of their peak response, and the amount of adaptation each exhibited to the same sustained stimulus. The fact that some units showed a sustained response to constant illumination stimuli of over eight minutes duration (cf. Fig. 2) strongly suggested that some amount of cupula displacement could be statically maintained. The fact that sustained cupula displacements were observed in traumatized preparations in response to a sustained caloric stimulus also supports the view that the thermal stimulus eventually produced quasi-steady convective torques on the ring of endolymph.

Lowenstein & Sand (1940) first reported a class of higher threshold units which are silent at rest, and which discharge only in response to rotation in one direction. We found that during excitatory caloric simulation, units of this type with progressively larger spike amplitudes were recruited. This finding is consistent with the observations of Taglietti et al. (1973) in the frog, who reported that afferent nerve spike size was related to threshold in a similar way. Taglietti and his collaborators suggested that, at least in the frog, the higher amplitude spikes may originate from larger fibers. However, only indirect evidence of this was offered. Although Erlanger & Gasser (1937) have reported that the amplitudes of nerve impulses recorded externally from peripheral nerves vary directly as the diameter of their nerve fibers, until this is convincingly demonstrated in the ampullary nerve of the skate, a relationship between fiber diameter and threshold must be regarded as tentative. Nonetheless, the existence of such a relationship in a non-mammalian vestibular organ containing only Type II hair cells is of some interest, since several authors have suggested that afferent response characteristics may be specified by a unit's innervation pattern in the crista and related characteristics such as fiber diameter as well as by the type and location of hair cell contacted (Walsh et al., 1972; Goldberg & Fernandez, 1977). The correlation of spike height and threshold observed in our preparation did not appear to be an artifact of the particular recording method used since it was also observed in similar experiments in which recordings were made from the undissected nerve using 20 μm tip diameter polished glass suction electrodes.

That a displacement of the top of the cupula was observed in response to caloric stimuli in cases where the cupula was first grossly stimulated validated the optical method. Since the caloric stimuli which produced this visible motion were identical to those which earlier produced no detectable motion, it appeared that the effective mechanical stiffness of the cupula had decreased as a result of the rapid dye injection. There are at least two possible explanations which might account for this change in stiffness: One possibility is that the cupula is normally adherent to the walls of the ampulla in a manner similar to that suggested by Hillman for the frog, and that injection of large amounts of fluid might overdeflect the cupula, or that (as suggested by Hillman, 1974) high concentrations of Alcian Blue dye might cause shrinkage of the cupula material, thus breaking its connection with the walls of the ampulla. On the other hand, there is as yet no direct evidence that the skate cu-

pula is attached to the ampullary walls, and on the basis of the present observations, it remains equally possible that the cupula is not so attached. Perhaps the jellylike aggregate in the ampulla seen after extensive staining normally contributes to cupula stiffness; agitation of such a gel might change its physical properties (cf. Dohlman, 1971; Grant & Van Buskirk, 1976).

Even with caloric stimuli sufficiently large to produce changes in single unit activity which approached 40 spikes/second, representing a four- or five-fold increase in activity in some cases, no cupula motion was observed. An argument that these stimuli correspond to relatively large angular acceleration stimuli can be made as follows. Data available for the skate (Lowenstein & Sand, 1940; Groen, Lowenstein & Vendrik, 1951) suggest that individual afferent units have acceleration sensitivities which lie in the range between 1 and 10 spikes/sec per deg/sec/sec. Most units described in the frog by Precht, Llinas & Clarke (1971) appear similar. Goldberg and Fernandez (1971) reported that units studied in the squirrel monkey varied in sensitivity between 0.5 and 4.0 spikes/sec per deg/sec/sec. Since the caloric stimulus can be considered physically equivalent to a sustained angular acceleration, we conclude that the larger stimuli used in the present experiments, which produced response changes up to 40 spikes/sec could be considered equivalent to sustained accelerations in the range of 4–40 deg/sec/sec. The majority of stimuli produced response changes of about 10 spikes/sec, equivalent to constant accelerations in the 1–10 deg/sec/sec range, continued for about 100 sec. Since these accelerations probably lie at the upper end or above the dynamic range of stimuli experienced by the animal in daily life, and yet no cupula motion was seen in response to equivalent caloric stimuli, it was concluded that the "normal" dynamic range of cupula motion in the skate is less than 3–5 μM, the resolution of the optical method used. If such a displacement represents an upper limit on the normal magnitude of cupula motion, it appears unlikely that cupula motion (be it bending, shearing, or deformation as a diaphragm) can be studied optically *in vivo* by these methods using stimuli of normal magnitude. Although potential species differences must, of course, be taken into account, we suggest that the results of previous experimental investigations in which the behavior of the semicircular canal cupula and/or afferent response was studied with cupula motions in excess of 5 μm should be interpreted cautiously (e.g. Steinhausen, 1927, 1931, 1933; Vilstrup, 1950; Ledoux, 1958; Trincker, 1962; Dohlman, 1960; Harada, 1972; Schmid et al., 1973; Taglietti et al., 1973; Flock & Goldstein, 1973; Valli et al., 1974; Grant & Van Buskirk, 1976; McLaren & Hillman, 1976). Our evidence suggests that the traditional view, perpetuated in many texts on vestibular function and anatomy, that the cupula normally moves as a swinging door within the ampulla over distances much greater than 5 μm is probably erroneous. Given that normal cupula motions appear to be only a very small fraction of the total dimension of the structure, it probably is more appropriate to describe the cupula as an enormously sensitive biological pressure transducer, one which apparently is easily traumatized by surgical overstimulation.

ZUSAMMENFASSUNG

Eine Höchstwertgrenze für das Ausmaß der Bewegung der Cupula wurde in dem isolierten Labyrinth des Skate experimentell bestimmt. Um die Cupula sichtbar zu machen, wurden, mit einer die ampullarische Wand durchstechenden Glaspipette, lokale Regionen der Cupula durch langsames Einflößen kleiner Mengen alcianblauer Tusche gefärbt. Man stellte fest, daß kalorische Reize, welche große Veränderungen bei Einzel-unit-activity in dem ampullarischen Nerv hervorriefen und welche öfters mehrere größere, bisher stumme Units anwarben, keine, durch die ampullarische Wand gesehen, nachweisbare Cupula-Bewegung hervorriefen. Allerdings wurde nach vorheriger starker Verschiebung der Cupula Bewegung als Reaktion identischer kalorischer Reize beobachtet. Die Analyse der afferenten Reizbeantwortungen weist darauf hin, daß die normale Reichweite der Cupula-Bewegung in diesem Tier unter der optischen Auflösung der Methode ist, die konservativ als 3–5 Mikrometer geschätzt wird.

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