MODIFICATION OF COCHLEAR MICROPHONICS AND ACTION POTENTIALS BY KCl SOLUTION AND BY DIRECT CURRENTS*

ICHIJI TASAKI AND CÉSAR FERNÁNDEZ
Central Institute for the Deaf, St. Louis, Missouri

EVER SINCE the time of Helmholtz, most physiologists have believed that different parts of the cochlea respond differently to various sound stimuli. Almost all the experimental evidence in support of this belief has been of rather indirect nature, however, and consequently there have been some differences of opinion among physiologists as to just how the behavior of the apical turn of the cochlea differs from that of the basal turn (see 8, 9). The differences might be resolved if we had a technique by which the responses of a part of the cochlea could be recorded, modified or eliminated without interfering with the responses of the remaining parts of the cochlea. The method of destroying the apical part of the cochlea has frequently been used by previous investigators, but it has several obvious disadvantages. For example, destruction of the apex may change the acoustic properties of the whole cochlea, the method cannot be applied to the other parts of the cochlea, and destruction of the apex may change the excitability of the sensory cells in the remaining parts of the cochlea.

In the present investigation we undertook to develop micro-techniques by which the condition of a part of the cochlea could be modified in a reversible manner. We have succeeded in recording electrical responses generated in any one of the turns of the cochlea without any significant contamination by the responses from other turns. Administration of an isotonic potassium chloride solution and application of direct current were chosen to modify locally the condition of the cochlea, because these agents are known to change reversibly both resting and action potentials in many other excitable tissues. Actually these agents were found to be very convenient for changing or totally suppressing these electrical signs of cochlear activity in a restricted region where the agents were applied.

The results show very clearly that the basal turn of the cochlea generates cochlear microphonics in response to both high- and low-frequency sound stimuli, whereas in the apical part high-frequency sound stimuli, applied through the external auditory meatus, do not give any response. Furthermore, the cochlear microphonics recorded from the round window of the guinea pig are the response of the basal turn and give practically no information about the activity of the more distant parts of the cochlea.

---

* This work was carried out under Contract N6onr-272 between the Office of Naval Research and Central Institute for the Deaf.

METHOD

Guinea pigs, anaesthetized with dial in urethane (0.5 cc./kg. body weight), were used for the experiments. Incision was made through the skin in the mandibular region, and both the masseter muscle and the mandible were cut across in the middle. Lifting the posterior part of the mandible with a retractor and pushing down the digastric muscle, the bulla was exposed. Then a part of the sterno-cleido-mastoid muscle was severed and the styloid process was cut across near its base. The surface of the bulla was then cleaned and, with a dental drill, an opening was made to expose the cochlea.

Recording electrodes were either enamel-insulated silver wire of approximately 100μ in diameter or nichrome-steel wire of about 20μ. The enamel was removed for approximately 100μ near the tip. This scraped tip was introduced into various parts of the cochlea

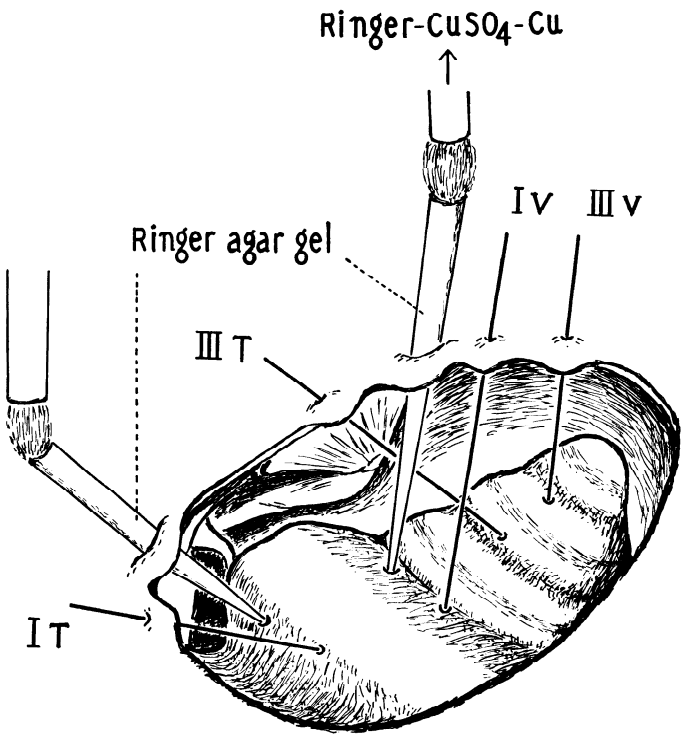

FIG. 1. Symbols I_V, I_T, II_V, II_T signify the wire electrodes leading to scala vestibuli of first turn, scala tympani of first turn, scala vestibuli of third turn and scala tympani of third turn respectively.

through small holes made with a very fine drill, sharpened under a dissecting microscope. Care was taken to avoid any injury in the scala media while making the holes and introducing the electrodes. The holes were just large enough to admit the electrodes and therefore the outflow of the perilymph through the hole (with the electrode in) was practically insignificant. The electrodes were fixed to the edge of the bulla with dental cement (Fig. 1). The animal was grounded through a clip on the neck.

The sound stimuli employed were either tone pips (4) or pure tones between 250 and 8000 c./sec. To obtain tone pips, rectangular voltage pulses were led to two sets of resonant circuits of the desired frequency; then the output was amplified and delivered to a loud speaker (Atlas PM-25). Pure tones were obtained from a beat-frequency oscillator (General Radio 1304-A), the transducer being the same as in the case of tone pips. The sound was generally delivered to the animal through a speculum fixed in the external

auditory meatus. The connection between the speculum and the loud speaker was made with a rubber hose 175 cm. long. Sometimes sound stimuli were delivered through the opening in the bulla.

Electrical responses from various parts of the cochlea were amplified with three independent channels of amplifiers (Grass) and were recorded simultaneously with three cathode-ray oscillographs. The input resistance of the amplifier was 0.5 megohm. Large condensers (1 μF) were inserted in the electrode circuit to block the D.C. component in the electrode circuit. For the responses arising in the basal turn, the method of electronic cancellation (3) was frequently used to separate action potentials and microphonics from each other.

For examining the effect of increased KCl in Ringer solution, a mixture of an isotonic (1.15 per cent) KCl solution and an ordinary mammalian Ringer solution (NaCl 9.0 g., KCl 0.42 g., $CaCl_2$ 0.24 g., and $NaHCO_3$ 0.2 g. made to 1 liter with distilled water) were prepared. A pair of small holes, each approximately 100μ in diameter, were made with a sharpened dental drill in the basal turn of the cochlea, one in the scala tympani and the other in the scala vestibuli. The testing fluid, generally stained with a small amount of neutral red, was introduced with a small pipette made of Pyrex glass into the cochlea through one of the holes in the basal turn. The fluid was seen to flow out immediately through the hole on the other side of the cochlear partition, undoubtedly passing through the helicotrema.

In the experiments in which elimination of the activity of the basal turn was desired, a pair of small holes was made in the scala tympani of the basal turn approximately 1.5 mm. apart. The isotonic KCl solution was introduced through one of these holes and escaped through the other. When an elimination of the function of only the upper turns of the cochlea was planned, just one hole, approximately 150μ in diameter, was made in the apex. The fluid was introduced into the cochlea with a fine glass pipette of approximately 50μ (outside diameter at the tip), injecting the fluid relatively deep inside the cochlea.

Direct current for polarization of the cochlea was applied through fine glass pipettes filled with Ringer-Agar gel connected to the Cu-$CuSo_4$-Ringer system (Fig. 1). The outside diameter of the tip of the pipettes was approximately 100μ. These pipettes, as well as the wire electrodes for recording, were fixed to the edge of the opening in the bulla with dental cement. The contact between the Ringer-Agar gel in the pipettes and the plastic tubes leading to the Cu-$CuSO_4$-Ringer systems was made with cotton wicks soaked in Ringer solution. A dry battery (up to 200 V.) was used as the source of current. Since the resistance of the glass pipette electrodes (approximately 300 kilohms each) was far greater than the resistance of the cochlear partition (of the order of 1 kilohm for D.C.), the strength of the polarizing current was determined simply by the voltage of the battery. Records of cochlear responses during passage of current were taken between 3 and 10 seconds after make of the current. The time from the start of the polarization did not significantly affect the size of the response.

RESULTS

1. Preliminary tests with applied potentials. A preliminary test with alternating currents revealed that the potential difference measured through the wire electrodes described above was slightly less than the difference in potential of the points in the fluid where the electrodes were immersed. For frequencies between 100 and 5000 c./sec., the loss in the observed potential was between 5 and 15 per cent with the silver electrodes and approximately three times larger with the nichrome-steel electrodes. The loss was greater both for lower and for higher frequencies and was independent of the A.C. voltage used for the testing (up to 2 mV.).*

Tests were also done to determine how far the current sent into the cochlea spreads along the cochlear partition. With a condenser-coupled

* (Added in proof) The use of a cathode-follower stage makes this loss in the recorded potential almost negligible (see Tasaki, Davis and Legouix, *J. acoust. Soc. Amer.*, 1952, *24*: 502–519).

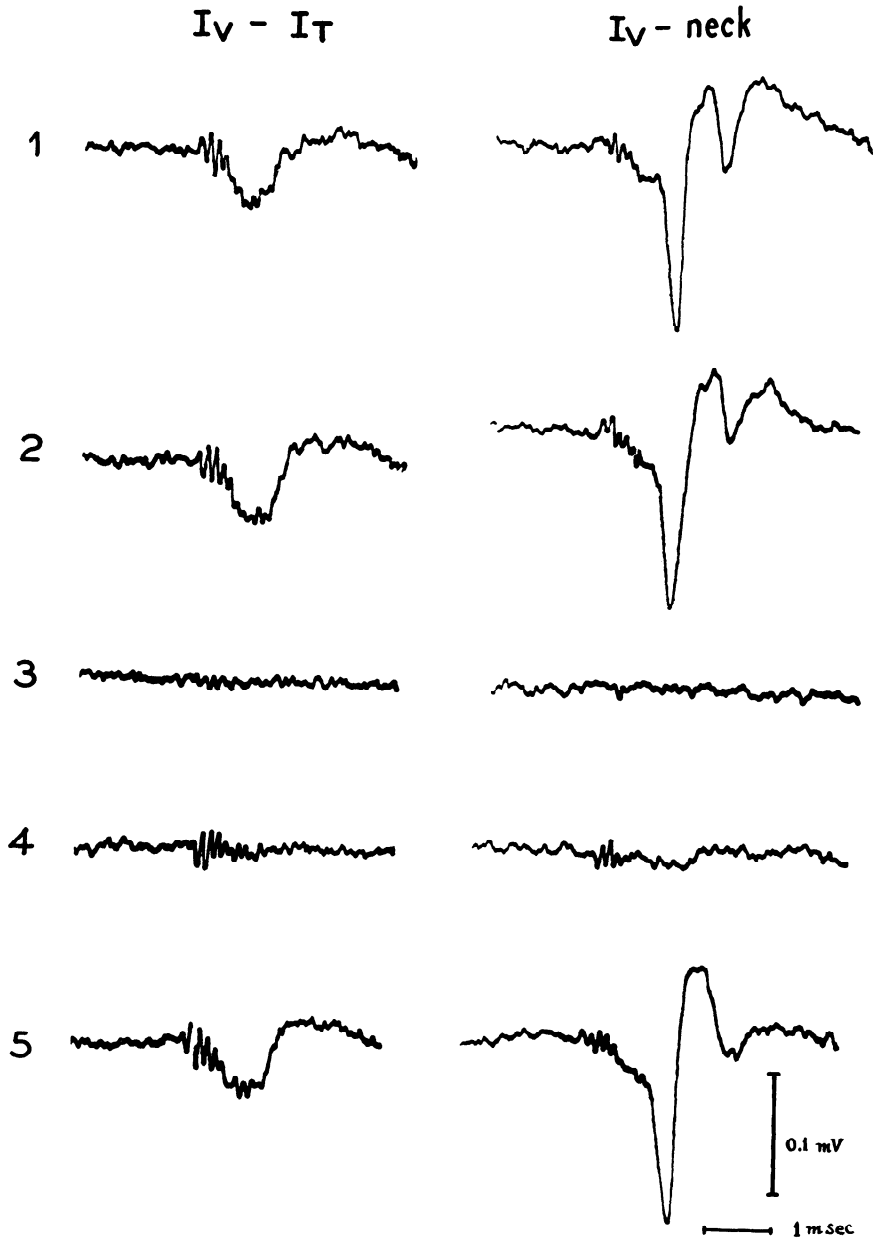

FIG. 2. Changes in cochlear responses caused by complete replacement of perilymph with Ringer solution and with mixture of one part of istonic KCl solution and four parts of mammalian Ringer solution. Stimulus: 8000 c./sec. tone pips at approximately 25 db above threshold for action potential. The uppermost records, 1, were taken before perfusion. Records 2, 8 minutes after replacement of perilymph with Ringer solution. Records 3, 4 minutes after introduction of the KCl mixture into cochlea. Soon after these records were taken, fluid in cochlea was replaced with normal Ringer solution. Records 4 were taken approximately 4 minutes after replacing KCl mixture with normal Ringer solution. Soon

amplifier and a cathode-ray oscillograph, measurements were made of the distribution of potential within the cochlea induced by means of a weak current (below 50 μA.) applied in the basal turn by the technique illustrated in Figure 1. When the glass pipette inserted in the scala vestibuli of the basal turn was connected to the source of current and the one in the scala tympani to the sink, the potential of the wire electrode in the scala vestibuli of the same turn (I_V in the diagram), referred to the wire electrode in the scala tympani (I_T), went up, the magnitude of this potential being proportional to the strength of the current applied. The potentials of the electrodes inserted in the upper parts of the cochlea were found to be between these two potential levels (I_V and I_T), but always closer to the potential of the scala tympani of the basal turn (I_T). No measurable potential difference was detected between the electrodes placed in the upper parts of the cochlea. This undoubtedly indicates that, with a source and a sink of equal strength placed across the cochlear partition of the basal turn, there is no spread of current into the upper part of the cochlea. Therefore the effect of direct current applied to the cochlea in this manner must be considered as the influence of current localized in the basal turn.

2. *Effect of KCl on cochlear responses.* In the first series of experiments an attempt was made to determine whether or not complete replacement of the perilymph in the cochlea with mammalian Ringer solution brings about any observable change in the excitability of the cochlea. First, with 8000 c./sec. tone pips, records were taken of the cochlear responses from the basal turn carrying a pair of recording electrodes and two holes for perfusion. Ringer solution was then introduced into the cochlea through a hole in the scala tympani and was allowed to flow out through the other hole in the scala vestibuli. The fluid accumulated in the bulla, but with a small pipette and small pieces of cotton wool the fluid was removed. As long as care was taken to remove this excess fluid, there was no detectable change in the threshold or the amplitude of the cochlear responses—both microphonics and action potentials—before or after replacement of the perilymph with normal Ringer solution.

In the next series of experiments the perilymph in the cochlea was completely replaced with mixtures of isotonic KCl solution and Ringer solution. The mixture was introduced just as in the preceding experiments. The surface of the cochlea was washed with fresh Ringer fluid and the fluid accumulated in the bulla was removed. After these procedures, which required 2–3 minutes, the cochlear responses—both the microphonics and the action potentials—were found to be reduced to an extent determined by the concentration of KCl in the mixture. With 8000 c./sec. pips at about 25

---

after this, cochlea was perfused with normal Ringer again. Records 5, approximately 4 minutes after second perfusion with normal Ringer solution. Records in left column show cochlear microphonic and so-called "summating potential." Those to right show also the two action potentials (recorded as downward spikes) that are usually known as "NI" and "NII."

db above the action potential threshold, the concentration of KCl in Ringer solution required to reduce the sizes of action potential to 50 per cent of its original value was approximately 0.15 per cent. With stronger sound stimuli a higher concentration of KCl was needed to obtain the same reduction of the response.

When the KCl solution was replaced later with fresh Ringer solution the cochlear responses recovered completely to their original size even when the concentration of KCl had been sufficiently high to suppress them almost completely (approximately 0.25 per cent KCl in Ringer; see Fig. 2). The recovery was much slower than the progress of the depression following application of KCl which seemed to reach a steady state within one minute. Furthermore, perfusion with normal Ringer solution had to be repeated several times before complete recovery was obtained. This probably is due to retention of some KCl in the scala media or in other parts of the cochlea where fresh Ringer solution could not reach directly.

After administration of pure isotonic KCl solution, washing inside the cochlea with fresh Ringer solution did not restore the action potential within one hour, although the cochlear microphonics still recovered fairly well.

3. Elimination by KCl of responses in upper or lower half of cochlea. When a small amount of isotonic KCl solution was introduced into the scala tympani of the basal turn, the responses recorded with electrodes inserted in the basal turn disappeared very rapidly. A similar treatment with KCl of the scala vestibuli eventually caused a total loss of responses from the basal turn, but the loss was definitely slower than when KCl was administered in the scala tympani. This slower action of KCl in the scala vestibuli may be due to slower diffusion of KCl through the tectorial membrane to the hair cells and nerve fibers or to possible dilution of KCl by cerebrospinal fluid entering by way of the cochlear aqueduct.*

At the moment when the response of the basal turn was eliminated completely by the action of KCl, there were still almost normal responses in the upper turns of the cochlea. Differential recording from the upper part of the cochlea was carried out in two ways. Sometimes one electrode was in scala vestibuli, the other in scala tympani of the third turn (see Fig. 3); sometimes both were in scala vestibuli, one in the third and the other in the apical turn. In all cases the responses from the upper turns were still almost normal in size at the moment when the response of the basal turn had just disappeared. Since a current with its source and sink in the cochlea has been shown not to spread to other turns of the cochlea, we believe that differential recording gives information only as to the response of the turn where the pair of electrodes is inserted. Actually, the response of the upper part of the cochlea as examined by this differential method behaved independently of the response of the basal turn.

* (Added in proof) A mixture of one part of an isotonic KCl solution and three parts of mammalian Ringer solution does not affect the cochlear responses at all when it is applied only from the vestibular side of the cochlear partition.

Differential recording from the upper turns, even without treatment with KCl, shows no response from these upper turns when high-frequency sound stimuli are applied. An 8000 c./sec. pure tone, which generates microphonics very effectively in the basal turn, is ineffective for the third and apical turns of the cochlea. With 8000 c./sec. tone pips, no microphonics of 8000 c./sec. were ever observed, but when the intensity of the 8000 c./sec. pip was high, slow microphonics, probably generated by the low-frequency components in the pip, were always observed.

Since the time when the cochlear responses were first discovered by

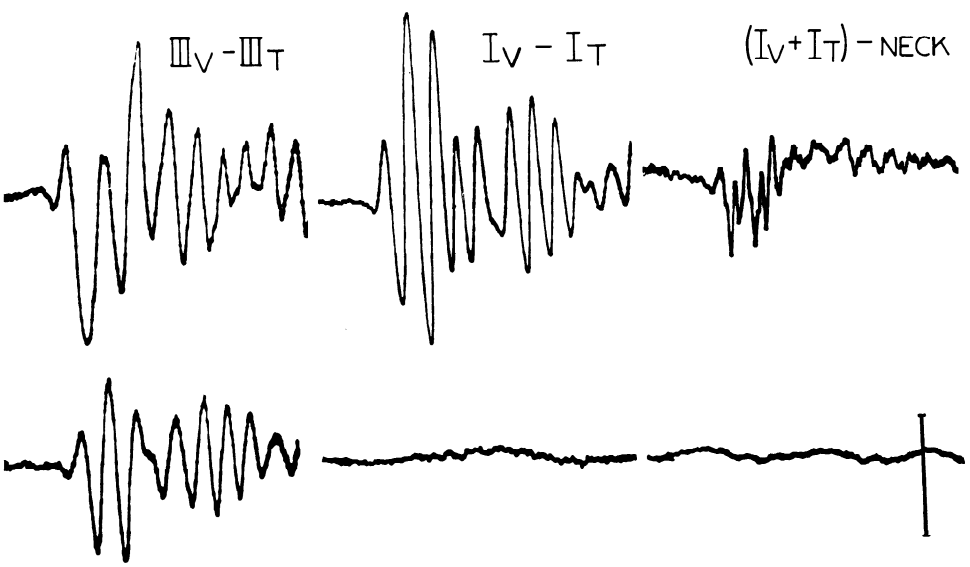

FIG. 3. Cochlear responses recorded before (top) and after (bottom) introduction of isotonic KCl solution into basal turn. Sound stimulus, 500 c./sec. tone pips at approximately 20 db above threshold for action potential. Placements of recording electrodes are indicated by symbols *III* (3rd turn), *I* (1st turn), *V* (scala vestibuli) and *T* (scala tympani). Records on right show almost pure action potentials obtained by method of electronic cancellation. Bar subtends 0.1 mV.

Wever and Bray (10) and were classified by Davis (2) into cochlear microphonics and action potentials, the region of the basal turn, including the region near the round window, has been known to respond to sound stimuli of varying frequencies. Now we have shown that the microphonics of the basal turn induced by low-frequency sound stimuli behave independently of the responses of the upper turns of the cochlea to the same stimuli. It is therefore evident that the low-frequency microphonics recorded from the basal turn by differential electrodes are actually the responses generated in the basal turn and not the spread toward the basal turn of microphonics generated in the upper part of the cochlea. In other words, with the sound stimuli applied through the external auditory meatus the region of the

cochlea near the round window generates cochlear microphonics in response to a sound of any audible frequency.

The function of the upper turns of the cochlea was next eliminated without affecting the response from the basal turn. The most successful method was to introduce a drop of KCl solution at the apex and wait until it spread down into the upper part of the cochlea. In addition to diffusion, periodic

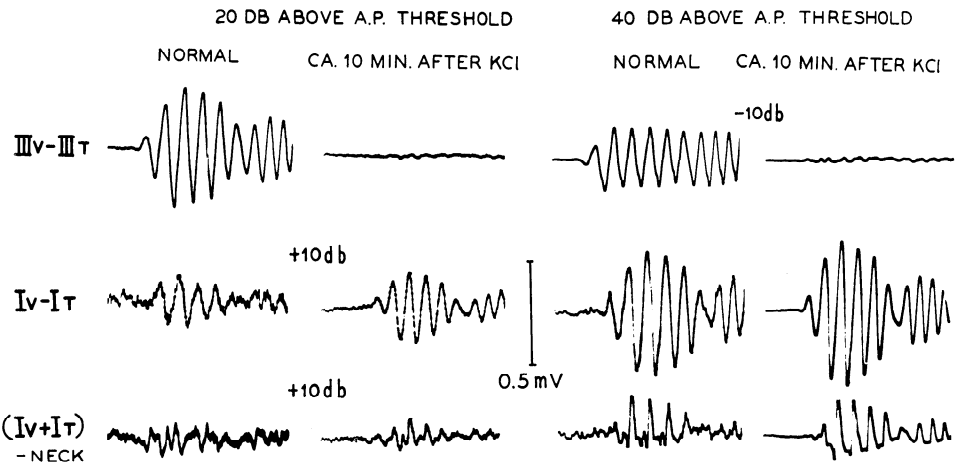

FIG. 4. Cochlear responses recorded before (1st and 3rd columns) and after (2nd and 4th columns) introduction of isotonic KCl solution into apex. Sound stimuli, 500 c./sec. tone pips at approximately 20 db (1st and 2nd columns) and 40 db (3rd and 4th columns) above threshold for action potential. Positions of recording electrodes and changes in sensitivity of amplifier are indicated in figure. Upper and middle records show cochlear microphonic; lower records show action potentials. Microphonic at turn *III* shows a different form with the stronger stimulus because at this intensity mechanism that generates microphonic is "overloaded" (9) and is nearing its maximum output. Therefore all of the waves have nearly same amplitude. Turn *I*, however, is not yet overloaded and microphonic still reproduces quite well the form of acoustic tone pip. Difference in form of tone pips from the two turns with strong stimulus is further evidence of independence of microphonics recorded from turns *I* and *III*.

movements of the stapes, resulting from contraction of the intra-aural muscles, seem to serve to bring the KCl downwards.

In Figure 4 is presented an example of such experiments. 500 c./sec. tone pips of two different intensities were used in this experiment. Responses from the basal turn were separated into microphonics and action potential by the method of electronic cancellation (3). The responses from the third turn were recorded simply by connecting the electrodes inserted in this turn to the two input terminals of a differential amplifier. Between 7 and 15 minutes after introduction of a drop of isotonic KCl solution into the apex, the response of the third turn was practically negligible, whereas the responses of the basal turn—both action potentials and microphonics—showed practically normal size during this period. Later, the response of the third turn began to recover and the basal turn responses became smaller,

apparently because of further spread and dilution of KCl in the cochlea. This experiment provides us with additional evidence that the responses recorded by the differential method with two electrodes placed in one turn is not significantly contaminated by the components generated in other turns of the cochlea.

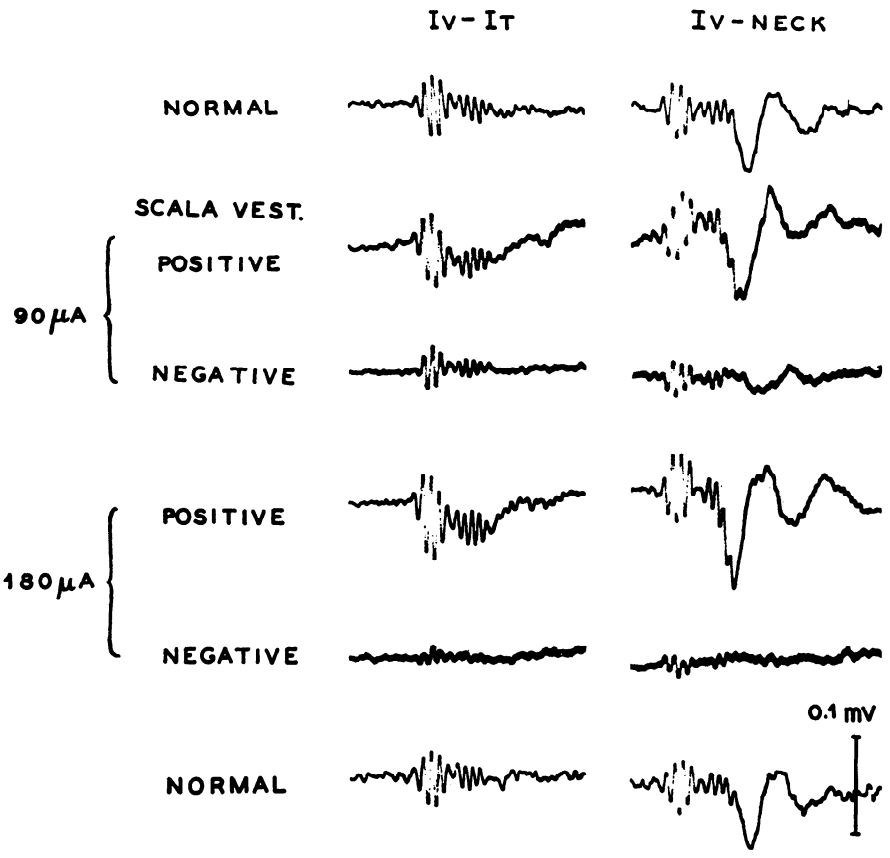

FIG. 5. Change in cochlear responses induced by D.C. applied in basal turn. Note also changes in latent period of action potential, which is shown in records in right column. Sound stimuli, 8000 c./sec. tone pips.

4. Effects of direct currents on cochlear responses. In this series of experiments, constant polarizing currents were applied to the basal turn through a pair of glass pipette electrodes and the responses were recorded with silver wire electrodes placed close to the polarizing electrodes. A D.C. microammeter and a series resistance of 0.5–1.0 megohm were generally connected in the battery circuit. Since the battery circuit was carefully isolated from the ground and since the resistance of this circuit was high as compared with that of the cochlea, there was no observable distortion or reduction in the responses because of the presence of the polarizing electrodes.

Figure 5 gives a typical example of the results obtained with 8000 c./sec.

tone pips. Without polarizing current, the potential of the scala vestibuli of the basal turn, referred to the neck, showed first a rapid vibratory excursion —namely, the cochlear microphonics—followed by action potentials. When a constant current of between 50 and 200 μA. was sent into the cochlea, both the microphonics and the action potentials were found to be modified in a perfectly reversible manner. With the source of current connected to the scala vestibuli and the sink to the scala tympani, the responses were enhanced and, with a current flowing in the opposite direction, depression of

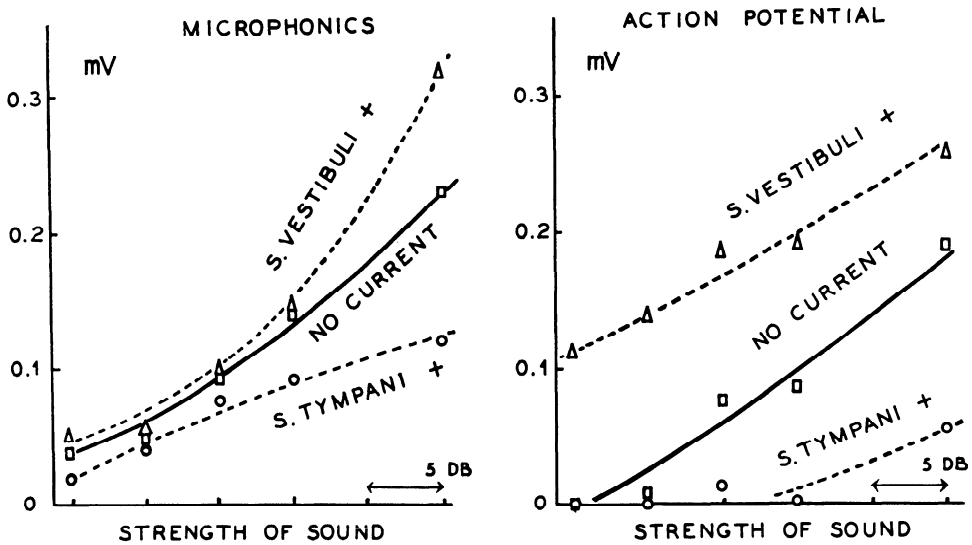

FIG. 6. Effect of D.C. applied in basal turn upon cochlear microphonics (left) and on action potentials (right) recorded from basal turn. Sound stimulus, 8000 c./sec. tone pips; strength of current, 150 μA.

the responses was observed. When the strength of the current was 200–350 μA. and its direction was such that the current traversed the cochlear partition from the scala tympani towards the scala vestibuli, the action potentials for 8000 c./sec. tone pips were in general completely suppressed by the current.* In an experiment with a fairly strong sound stimulus and with currents of varying intensities, an approximate parallelism was observed between the change in the action potential and that in the cochlear microphonic. The effect of polarizing currents upon the microphonic was more marked when the sound stimulus was strong than when it was near the threshold (Fig. 6).

The effects of D.C. upon the cochlear microphonics induced by pure tones were also investigated with similar arrangements of the electrode. Again, the current flowing from the scala vestibuli towards the scala tym-

* Further increase in current caused an irreversible change in the cochlea. No reversal in the sign of the microphonic was observed by D.C. polarization.

pani increased the amplitude of the microphonics and the current flowing in the opposite direction brought about an opposite effect. The depressive or enhancing effect of the current increased with the intensity of the current. Within limits the change in the amplitude of cochlear microphonic was found to increase approximately directly with the current strength (see Fig. 7). The depressive effect was in many cases more marked than the enhancing effect. These effects of polarization did not seem to increase with time during passage of a constant current, but a postpressive enhance-

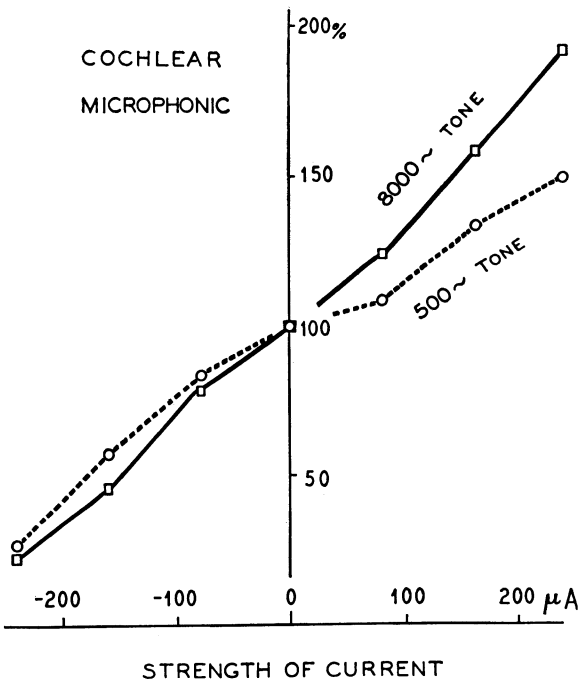

FIG. 7. Effect of D.C. upon cochlear microphonics induced by pure tones. Current was applied in basal turn and responses were recorded from same turn. Strength of sound was fixed at value which just gave maximum cochlear microphonic when there was no D.C. Maximum microphonic was 0.42 mV. for 8000 c./sec. tone and 1.1 mV. for 500 c./sec. tone. Negative value of current intensity signifies that current flowed through cochlear partition from scala tympani toward scala vestibuli.

ment was often observed during the period 3–5 sec. after cessation of the current.

Figure 7 gives an example of the results showing the effect of direct current as a function of current intensity. The intensity of the sound stimulus was in this case such that the amplitude of the microphonic, observed without applied current, was maximal. The current affected the responses to both 500 and 8000 c./sec. pure tones, but the effect on the lower tone was

slightly less than that on the higher tone. The effect on an intermediate tone (2000 c./sec.) was also tested on this animal; the result was similar to those for the other frequencies.

5. Local modification of cochlear response by direct current. According to what has been stated up to this time, the polarizing current applied in the basal turn should not affect in any way the response of the upper turns of the cochlea. It has been shown that the current applied in the basal turn does not spread toward the upper turns of the cochlea. It has also been demonstrated that the method of differential recording of the cochlear microphonic from any turn gives information as to the activity of that turn without contamination by the responses of other turns. If, therefore, the microphonics from the basal and third turns are recorded simultaneously, the polarizing current, applied in the basal turn, should change the amplitude of the response from the basal turn and leave the response from the third turn unaffected. Figure 8 shows that this is actually so. The arrangement of the electrodes for this experiment is given in Figure 1. In order to evoke cochlear microphonics of appreciable size in the third turn, low-frequency sound stimuli—pips in this case—were used. The current applied was very strong, strong enough to suppress the response of the basal turn when it was flowing through the cochlear partition from the scala tympani toward the scala vestibuli. During the passage of this current in the opposite direction, there was an increase of approximately 50 per cent in the amplitude of the microphonics from the basal turn. The response from the third turn remained absolutely unchanged during and after passage of the current.

The question now arises: how far does the effect of a polarizing current spread from the site of application along the cochlear partition? A rather crude experiment with a weak constant current indicated that the decay of D.C. potential across the cochlear partition was something like 6 db/mm. within the basal turn. The total length of the basal turn of the guinea pig cochlea is known to be about 8 mm. Therefore, with a source and sink of current located across the cochlear partition near the round window, the density of current in the second turn should be less than 1 per cent of the value near the round window. Cochlear microphonics generated by low-frequency sound stimuli seem to spread along the cochlear partition in the same manner as the applied constant current. In the upper turns, where the basilar membrane is wider and the two scalae are smaller, spread of both applied current and microphonics is probably far less than in the basal turn.

Discussion

Many of the implications of our results have been pointed out in the previous section as the results were described. There are, however, several more general aspects.

Several previous investigators have employed the cochlear microphonic for the study of the localization of mechanical activity within the cochlea as

a function of frequency. Probably all of them assumed that the microphonic as recorded between the round window and a reference electrode on the neck gave some sort of information as to the activity of the entire cochlea. Actually this is not the case. Figure 3 shows that the microphonic generated

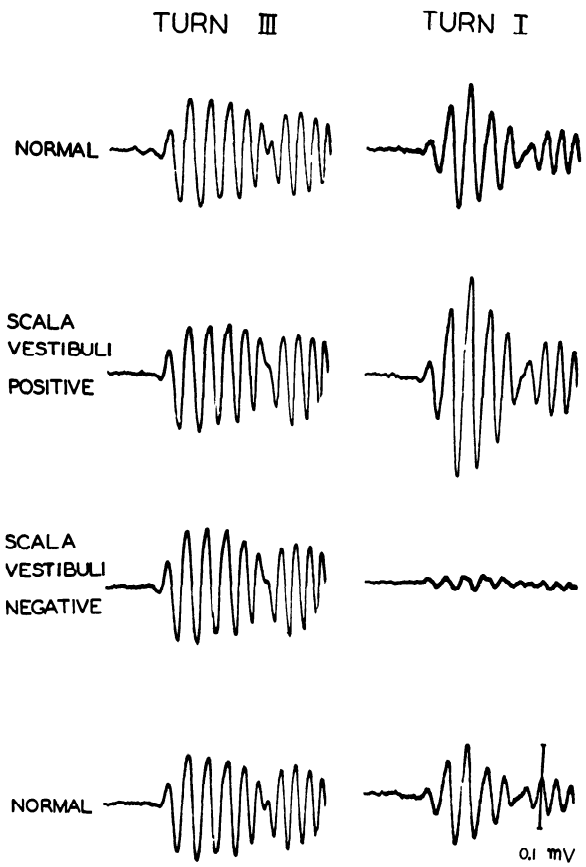

FIG. 8. Effect of D.C. applied in basal turn upon cochlear microphonics recorded from basal (right) and third (left) turns. Sound, 500 c./sec. tone pips as approximately 30 db above threshold for action potential. Current was 0.3 mA. Difference in shape of responses from turns *I* and *III* respectively is due to earlier "overloading" of cochlea in turn III when stimulus is 500 c./sec. tone pip. (See legend of Fig. 4.)

in the upper parts of the cochlea does not appreciably affect the potential recorded from the basal part. The contribution from the upper turns cannot be more than about 3 per cent at most.

It was recognized by previous investigators that the microphonic generated at the apex but recorded from the round window would be "attenuated" considerably. Davis *et al.* (3), for example, estimated this attenuation as approximately 30 db. But this attenuation is far less than our present

estimate of approximately 6 db/mm., or at least 100 db from the uppermost turn to the base. It should be emphasized, however, that there is an important innovation in method in the present experiments. We have employed two electrodes inserted into a given turn of the cochlea, one in scala vestibuli and the other in scala tympani. Davis *et al.* inserted one electrode into scala vestibuli or apex but used an electrode in the neck of the animal as an "indifferent" reference point.

The complication caused by using the neck as a reference point is clarified by the following observation: at the moment when the activity of the third and apical turns of the cochlea was eliminated by local application of KCl (experiment of Fig. 4) we recorded the potential difference between one of the electrodes in the third turn and the ground electrode on the neck and found that there was still a fairly large microphonic. This microphonic, which undoubtedly was produced by the activity in the lower turns of the cochlea, was frequently larger than the normal response of the third turn recorded by the differential method. From this and several other observations, we could demonstrate that the microphonic recorded with one electrode in the *apex* and the other on the neck was a mixture of responses from all the turns of the cochlea.

For the analysis of the responses from the *basal turn*, however, there is no complication of this type. The microphonic recorded with one electrode on the neck and the other in the basal turn, either in scala vestibuli or in scala tympani, gives information as to the activity taking place in the basal turn without contamination by the responses originating in the upper turns (Figs. 3, 8). It is therefore possible to separate the action potentials from the microphonics of this turn very completely by the method of electronic cancellation. If the neck electrode was not truly indifferent but was being affected by the microphonic generated in the apical region, this potential coming from the apical parts would remain, like the action potential, when the I_V-neck and I_T-neck potentials are added to cancel out the local microphonics. (The cancellation occurs because the microphonic potentials in I_V and I_T are opposite in phase.)

It is clear from our present observations that a D.C. potential difference applied in the basal turn between scala vestibuli and scala tympani does not cause any observable potential difference across the cochlear partition in the second, third or apical turns. The rapid attenuation of this potential difference can readily be understood if we assume that the bony walls of the cochlear canal of a living animal have a high electrical resistance. Perilymph is a relatively good conductor of electricity with specific resistance of less than 100 ohm-cm. Impedance measurements made recently in this Institute (which will be published elsewhere) show that the normal cochlear partition has a fairly high resistance to direct currents. The relationships between the conductivity of the cochlear partitions, the perilymph and the bony wall of the canal allow some spread of potential difference along the basilar membrane, but the attenuation is apparently of the order of 6 db/mm. in the basal turn.

In spite of the attenuation of potential difference between the two sides of the basilar membrane, we find that microphonics in response to a 500 or 250 c./sec. tone can be recorded between scala vestibuli and scala tympani of the basal turn. We must therefore conclude that the basal turn, as well as the higher turns, can generate cochlear responses when low-frequency sound is applied to the ear. Our direct tests with KCl and D.C. confirm this conclusion.

Our experiments also show that the third and apical turns do *not* respond to high-frequency sounds. This unsymmetrical situation seems qualitatively consistent with the direct observations by Békésy (1) of the movements of the cochlear partition of the dead ear in man and animals. It is our present impression, however, that the cochlear microphonics recorded from the basal turn are larger than we should expect from Békésy's resonance curves which show a fairly rapid decay in amplitude of vibration of the cochlear partition towards the round window.

The present experiments show that the cochlear microphonic can be profoundly modified by KCl and by D.C. polarization. Very probably these agents modify the microphonic by changing the resting potential of the cells in the organ of Corti. The observations therefore support the hypothesis that the microphonic depends on the membrane potential of the organ of Corti.

Our observation on the effect of D.C. upon the size of the action potential reminds us of similar observations on other sensory endings (on the muscle spindle, 7; on the eye, 5; on the Pacinian corpuscle, 6; on the tactile ending in the frog skin, Tasaki, unpublished). All of these observations demonstrate strong interactions between the effect of sensory stimuli and electric currents applied to the region of the sensory nerve endings. We believe that in our experiments the applied current affects the process of initiation of propagated nerve impulses in the sensory nerve fibers and not the processes of conduction. It is unlikely that the D.C. depresses the size of action potential by blocking the nervous conduction in the sensory nerve fibers because the effect of D.C. is very clear at current strength of below 50 μA. which generates a potential difference of less than 50 mV. across the cochlear partitions. It is well known that for an anodal block of conduction in small fibers much higher voltages are needed. Furthermore, it is impossible to interpret the increase in the size of the action potential by D.C. on the assumption that D.C. affects the nervous conduction.

Summary

1. Techniques were developed to record cochlear responses from different turns of the guinea pig cochlea simultaneously, to perfuse the cochlea with chemicals, and to polarize the sensory endings in the cochlea by D.C.

2. Complete substitution of the perilymph with normal mammalian Ringer solution does not cause any change in the electrical responses of the cochlea to sound stimuli.

3. An increase in KCl content in the perfusing fluid reduces cochlear microphonic and action potentials reversibly.

4. Elimination of the responses of the apical turn by isotonic KCl solution does not affect the responses of the basal turn. Complete suppression of the responses of the basal turn by KCl or D.C. does not influence the cochlear microphonic of the third turn.

5. The basal turn of the cochlea responds to high-, to middle- and to low-frequency sounds. The third and apical turns respond only to low-frequency sounds.

6. A direct current traversing the cochlear partition from the scala vestibuli (source) to the scala tympani (sink) enhances the cochlear responses, both cochlear microphonic and action potential. A current flowing in the opposite direction brings about depression of both responses.

ACKNOWLEDGMENT

The authors wish to express their appreciation of the helpful advice and criticism given by Dr. Hallowell Davis in this work.

REFERENCES

1. BÉKÉSY, G. V. The vibration of the cochlear partition in the anatomical preparation and in models of the inner ear. *J. acoust. Soc. Amer.*, 1949, *21:* 233–245.
2. DAVIS, H. The physiological phenomena of audition. Pp. 962–986 in: MURCHISON, C. A. *Handbook of general experimental psychology.* Worcester, Mass., Clark Univ. Press, 1934.
3. DAVIS, H., FERNÁNDEZ, C., AND MCAULIFFE, D. R. The excitatory process in the cochlea. *Proc. nat. Acad. Sci., Wash.*, 1950, *36:* 580–587.
4. DAVIS, H., SILVERMAN, S. R., AND MCAULIFFE, D. R. Some observations on pitch and frequency. *J. acoust. Soc. Amer.*, 1951, *23:* 40–42.
5. GRANIT, R. AND HELME, T. Changes in retinal excitability due to polarization and some observations on the relation between the processes in retina and nerve. *J. Neurophysiol.*, 1939, *2:* 556–565.
6. GRAY, J. A. B. AND MALCOLM, J. L. Initiation of nerve impulses by mesenteric Pacinian corpuscles. *Proc. roy. Soc.*, 1950, *137B:* 96–114.
7. MATTHEWS, B. H. C. The response of a single endorgan. *J. Physiol.*, 1931, *71:* 64–110.
8. STEVENS, S. S. AND DAVIS, H. *Hearing: its psychology and physiology.* New York, John Wiley and Sons, 1938. xiii, 472 pp.
9. WEVER, E. G. *Theory of hearing.* New York, John Wiley and Sons, 1949. xiii, 484 pp.
10. WEVER, E. G. AND BRAY, C. W. Auditory nerve impulses. *Science*, 1930, *71:* 215.