WADD TECHNICAL REPORT 60-778

ORGAN OF CORTI'S NEGATIVE INTRACELLULAR POLARIZATION

Edward A. Rice, Capt., USAF

Biomedical Laboratory
Aerospace Medical Laboratory

JANUARY 1961

Project No. 7232
Task No. 71789

WRIGHT AIR DEVELOPMENT DIVISION
AIR RESEARCH AND DEVELOPMENT COMMAND
UNITED STATES AIR FORCE
WRIGHT-PATTERSON AIR FORCE BASE, OHIO

900 – May 1961 – 27-994

FOREWORD

This investigation was performed by the Bioacoustics Branch, Aerospace Medical Laboratory, Wright Air Development Division, in support of Project No. 7232, "Research on the Logical Structure and Function of the Nervous System," Task No. 71789, "Physical and Physiological Mechanism Involved in Reception of Acoustic Energy."

The author expresses his appreciation to Dr. Joseph R. Mundie for his comments and recommendations during this investigation and to Dr. David H. Hubel for his valuable assistance in preparing the tungsten electrode.

Animal experimentation was performed in accordance with the rules for animal care as established by the American Medical Association.

ABSTRACT

The negative potential located between the basilar membrane and the reticular lamina of the guinea pigs was investigated using several types of electrodes from 3 to 24 microns in diameter. This potential was found intracellular in origin and located within the organ of Corti. The hypothesis that the negative potential found in the scala media of hypoxic animals represents a "leakage" of the organ of Corti's negative intracellular polarization is supported by these experiments. The negative intracellular polarization of the organ of Corti was found to exist during the life span of the microphonics. The significance of these findings in explaining the origin of microphonics is discussed.

PUBLICATION REVIEW

E. L. deWILTON, CAPT., MC, USN
Acting Chief, Biomedical Laboratory
Aerospace Medical Laboratory

ORGAN OF CORTI'S NEGATIVE INTRACELLULAR POLARIZATION

INTRODUCTION

Recent investigation (ref. 5) has revealed the presence of a negative potential between the scala media and scala tympani of hypoxic animals. We undertook the following study to determine if the negative intracellular polarization of the organ of Corti is the source of this potential. We also wanted to determine if the organ of Corti's negative intracellular polarization is in existence during the life span of the cochlear microphonics. This is a necessary requirement before this potential can be considered as a source of energy for the "biological amplifier" in the variable resistance theory of the cochlear microphonics (refs. 2, 3).

METHOD AND RESULTS

The instrumentation is illustrated in figure 1.

Location of the Negative Intracellular Polarization

We devised a method to determine the location of the electrode tip as it penetrated the basilar membrane and the organ of Corti. A Bak preamplifier (ref. 1) was used to record changes in the impedance of the electrode with reference to ground as it traveled from scala tympani to scala media through the basilar membrane and the organ of Corti. A transducer was attached to the micromanipulator to permit the depth of penetration to be displayed on a cathode ray tube. A slight change was noted in the electrode's impedance on intial contact with the basilar membrane. When the electrode had traveled 10 microns beyond the initial contact point, the dc potential suddenly decreased from zero to a negative 60 to 75 mv (figure 2). This negative potential reappeared in some experiments on withdrawal of the electrode. In some experiments the negative potential persisted until the electrode reached a position where a much larger impedance change took place. Since there was a reversal in the phase of the microphonics after the large impedance change, we thought the change was caused by the electrode penetrating the reticular lamina. The distance from the basilar membrane to the reticular lamina measured approximately 55 microns. In other experiments the negative potential disappeared 5 to 10 microns beyond the point where it was initially recorded.

We determined the extent of negative potential relative to the basilar membrane's width by penetrating with the electrode at selected points along the width of the basilar membrane. We removed the round window membrane and aspirated the perilymph in the scala tympani. A negative potential from 60 to 75 mv was recorded from all points across the basilar membrane. Distribution of the negative potential in depth was constant regardless of the point of penetration.

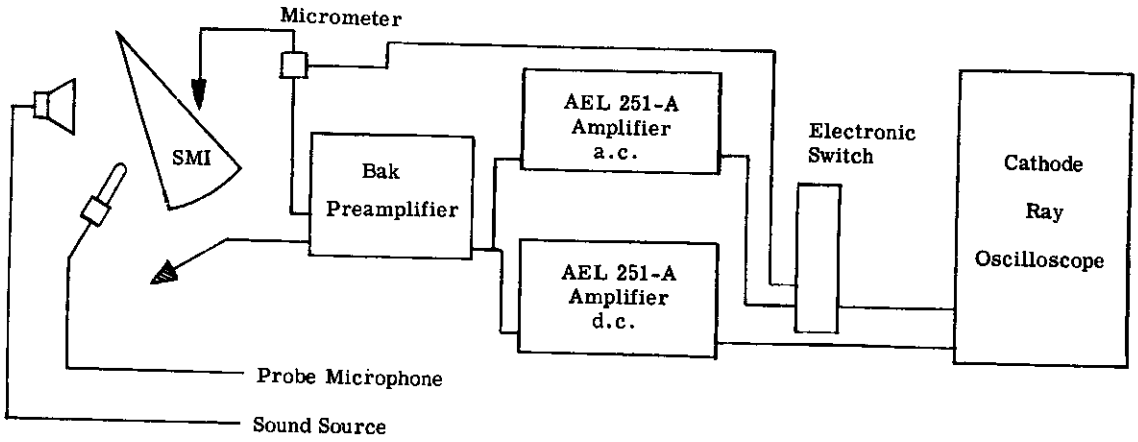

Figure 1. Schematic Illustration of the Instrumentation

The Negative Intracellular Polarization

The maximum recording time of the potential was approximately 2.5 minutes. We thought the short period of time the negative potential could be recorded was due to poisoning of the source from diffusion of electrolyte, inability to stabilize the electrode on the membrane, or damage to the source as a result of using electrodes with improper tip size. We used several approaches to investigate the cause of the short recording period of the negative potential.

In the first approach metal electrodes were substituted for the glass pipette electrodes. The metal electrode was an electrolytically sharpened tungsten wire (ref. 4). Metal electrodes eliminated the possibility of poisoning of the generating mechanism as a result of the electrolyte diffusing out of the electrode. Metal electrodes did not prolong the recording time of the potential.

In the second approach a metal electrode was used with a shoulder 20 microns from the tip. The 100- to 150-micron shoulder stabilized the electrode by preventing a backlash movement of the basilar membrane. We prepared the electrode by inserting an uncoated piece of electrolytically sharpened tungsten wire into a 1-mm (OD) glass pipette. The pipette was placed over a small flame and the sharpened end of the tungsten wire slowly pulled through the melted glass. The glass-coated tip was reinserted into the flame and the glass formed a ball a few microns from the tip. The distance from the tip to the glass shoulder was controlled by regulating the time the glass-coated tungsten tip was held over the flame. An electrode with a shoulder 20 microns from the tip recorded a negative potential of 60 to 75 mv and did not prolong the duration of the negative potential. The shoulder prohibited the electrode from penetrating into the scala media.

We used electrodes with tips of various diameters. Regardless of tip size the maximum duration in which the negative potential could be recorded was 2.5 minutes. At the end of this period the potential rapidly returned to zero.

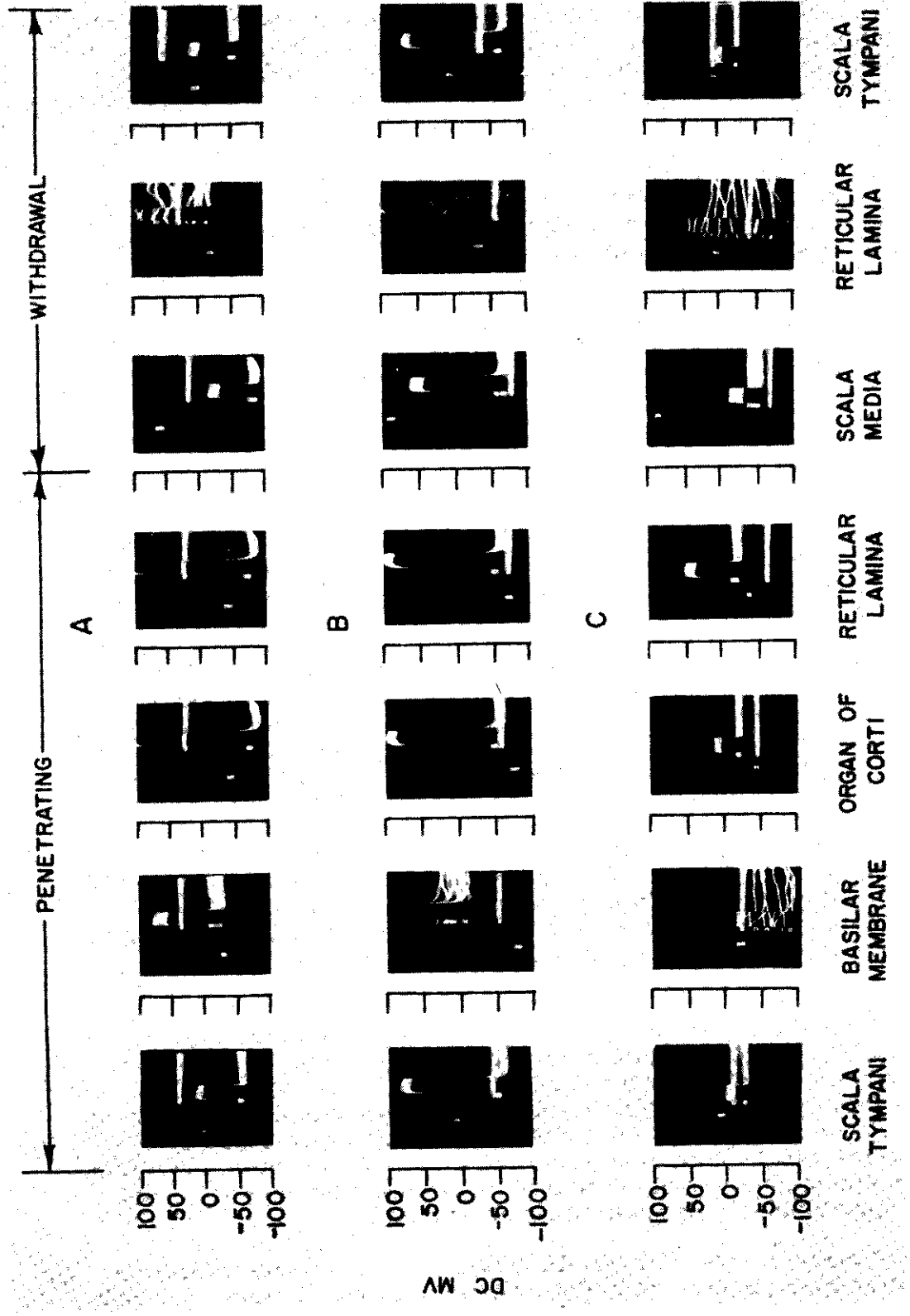

Figure 2. Penetration of the Basilar Membrane and Organ of Corti Showing the Electrode's Impedance and DC Potential Changes During Penetration

The DC potential is represented by the dot. The horizontal bar represents the depth of penetration (calibration: A=1 line/42.8, B and C=1 line/25.2). The amplitude of the deflection of the third trace represents the electrode resistance and the capacitance changes can be interpreted from the shape. Note the negative potential on withdrawal.

The Effects of Oxygen Deprivation on the Negative Intracellular Polarization

The short duration of the negative potential precluded obtaining a complete hypoxic curve on each animal. Repeating penetrations in the same animal decreased the magnitude of the negative intracellular potential. Thirty guinea pigs were divided into 6 groups. Group A was used as control (no nitrogen). In the remaining five groups penetrations were made at controlled intervals after hypoxia (figure 3). In group B penetrations were made 5 minutes after hypoxia. There was no change in the negative intracellular polarization. However, the scala media was negative in respect to the scala tympani. The microphonics decreased but could still be detected. In group C penetrations were made 30 minutes after hypoxia. There was no change in the negative intracellular polarization. It was still in the 60- to 75-mv range. Also, the scala media's negative potential remained 30 to 35 mv. In group D, 1 hour after hypoxia, both potentials had decreased. The negative intracellular polarization was 40 mv and the negative potential of the scala media was 20 mv. In group E, 2 hours after hypoxia, the negative intracellular polarization had decreased to 35 mv and the scala media was 10 mv. Five hours after cessation of the animal's oxygen supply, both negative potentials and microphonics had disappeared.

DISCUSSION

When viewing the negative intracellular polarization of the organ of Corti as a possible source of the scala media's negative potential, particular consideration should be given to the evidence presented on the effects of oxygen deprivation. The parallel persistance and final decay of both potentials support the hypothesis that the scala media's negative potential represents a leakage of the negative intracellular polarization of cells in the organ of Corti.

However, one important question remains unanswered: is the leakage due to the effect of oxygen deprivation or is it present in normal animals? If it is a result of hypoxia, then it may be regarded as an artifact. If, however, it is present in normal animals, then the implication is of great importance. If the resistance of the reticular lamina varies with its deformation, then the potential of the scala media would become less positive as the negative potential leaked through. The magnitude of the positive potential of the scala media would depend on the electrical resistance of the reticular lamina. This, in turn, would be determined by the degree of deformation. Leakage of the negative potential from the organ of Corti as a result of deformation of the reticular lamina would also cause the organ of Corti to be less negative. Thus, an electrode placed on one side of the reticular lamina would record an alternating potential opposite in phase to an alternating potential recorded on the other side of the membrane in response to sound.

The results of experiments—those in which destruction of the basilar membrane was accomplished within 20 microns of the point of penetration, those in which distribution of the negative potential was measured along the width of the basilar membrane, and those in which the distribution of the negative potential was measured in depth—clearly indicate that the negative potential of the organ of Corti is locally produced by cells. This conclusion agrees with the findings of others (refs. 6, 7, 8). However, the results of the experiments with large electrodes show that penetration of the cells with small electrodes is not a prerequisite to demonstrating this potential.

As a source of energy for the "biological amplifier" in the variable resistance theory of the cochlear microphonics, the negative intracellular polarization of the cells on the organ of Corti meets the requirement that the negative potential must exist for as long as the microphonics.

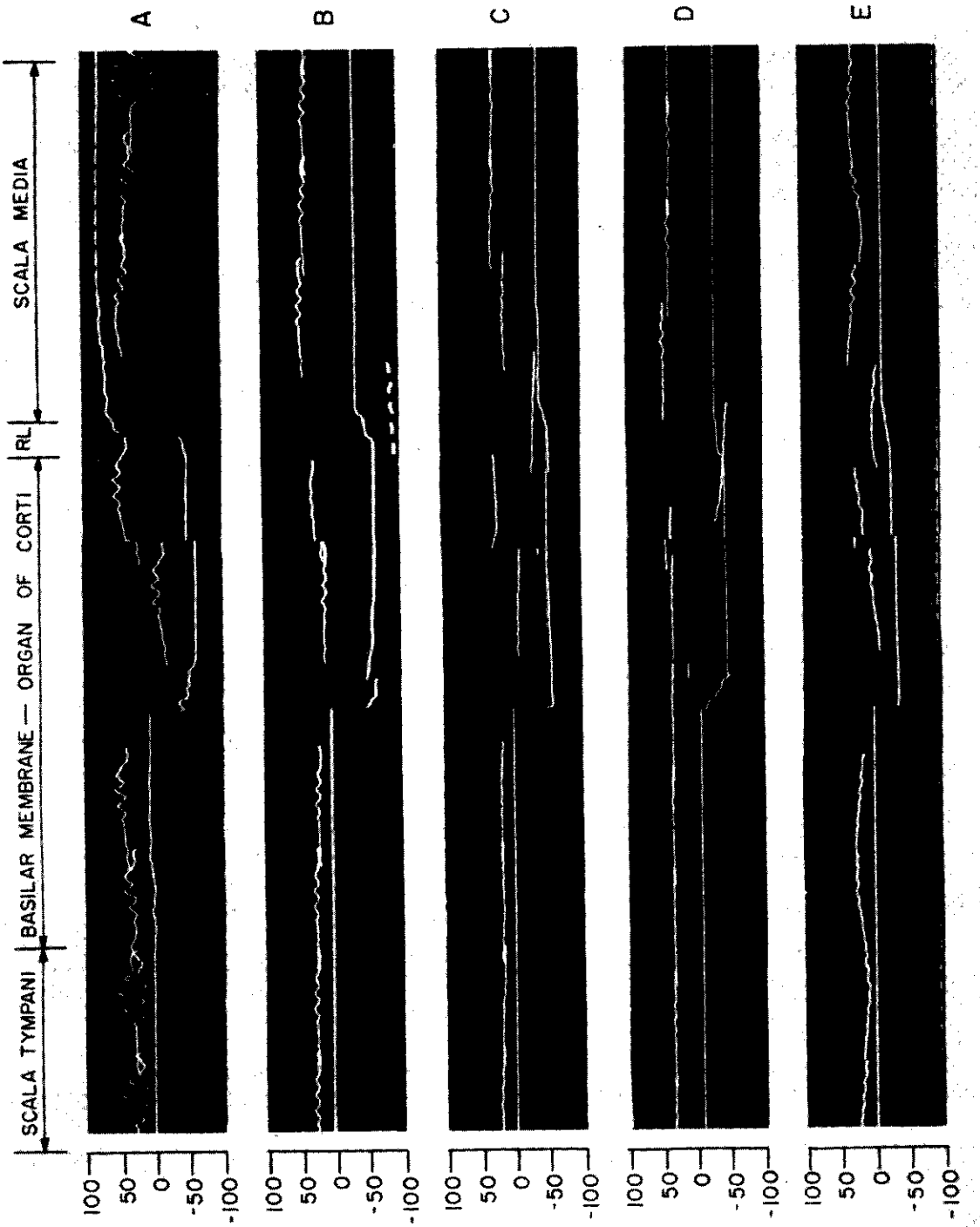

Figure 3. Postmortem Deteriation of the Organ of Corti's Negative Intracellular Polarization

Each line represents a separate penetration from scala tympani to scala media. A=Control, B=5 minutes after hypoxia, C=30 minutes after hypoxia, D=1 hour after hypoxia, and E=2 hours after hypoxia.

SUMMARY

The negative potential, located between the basilar membrane and the reticular lamina, was investigated using various types of both metal and glass pipette electrodes.

This negative potential was found intracellular in origin. However, demonstration of this potential does not require penetration of the cells with fine electrodes. In this investigation the negative potential was recorded with electrodes 24 microns in diameter.

The results of the investigation support the hypothesis that the scala media's negative potential observed in hypoxic animals represents a leakage of the negative intracellular polarization of the organ of Corti into the scala media.

The negative intracellular polarization was found to exist during the life span of the microphonics.

BIBLIOGRAPHY

1. Bak, A.F., "A Unity Gain Cathode Follower," EEG and Clin. Neurophysiol., Vol 10, p. 745, 1958.

2. Davis, H., "Energy into Nerve Impulse: Hearing," Med. Bull. St. Louis Univ., Vol 5, p. 43, 1953.

3. Davis, H., "Mechanism of Hearing," Nerve Impulse, Progress Associates, Caldwell, New Jersey, 1953.

4. Hubel, D.H., "Tungsten Microelectrode," Science, Vol 12, p. 549, 1957.

5. Rice, E.A., and E.W. Shinabarger, Studies on the Endolymphatic DC Potential of the Guinea Pig's Cochlea, WADD Technical Report 60-162, Wright Air Development Division, Wright-Patterson Air Force Base, Ohio, 1960.

6. Tasaki, I., H. Davis, and D.H. Eldredge, "Exploration of Cochlea Potentials in Guinea Pig with a Microelectrode," J. Acoust. Soc. Am., Vol 26, p. 765, 1954.

7. von Békésy, G., "DC Resting Potentials Inside the Cochlear Partition," J. Acoust. Soc. Am., Vol 24, p. 72, 1952.

8. von Békésy, G., "Gross Localization of the Place of Origin of the Cochlear Microphonics," J. Acoust. Soc. Am., Vol 24, 409, 1952.