Patella vulgata, *Nomaeopelta dalliana*, *Lottia gigantea* (Fig. 1*B*), and *Acmaea mitra* (Fig. 1*C*) check those of the goethite sample (Fig. 1*A*) in both spacing and intensity of lines.

This is the first indication that goethite is precipitated by marine invertebrates. After death of the animals, the goethite of the denticles should be incorporated into the marine sediments. Whether goethite is stable in any of the sedimentary environments of the sea remains to be determined.

Of the gastropods investigated here, the species of *Acmaea*, *Patella*, *Nomaeopelta*, *Lottia*, *Fissurella*, *Diadora*, *Nerita*, *Tegula*, and *Littorina* live on rocky surfaces in the littoral zone and on rocks at shallow depth in the euphotic zone. These species feed on filamentous algae attached to rocks. It has been shown, particularly for limestones, that the filamentous algae enmesh the surfaces and commonly also bore into the rocks to a depth of a few millimeters (*7, 8*). The boring algae cause weakening and, frequently, disintegration of the rocks. Gastropods and chitons which feed on filamentous algae remove the filaments from the rocks by scraping with their radular teeth. Some species of gastropods are reported to graze only on the filaments attached to the surface, whereas other gastropod species, such as the chitons, are able to feed also on embedded algae by penetrating the rocks with their radular teeth (*2, 7–9*). There is uncertainty whether some gastropods can remove boring algae when they are embedded in consolidated rocks or only when the rock particles are mechanically weakened by the boring algae (*8*), and it is important to know whether algae-feeding gastropods are capable of effective biomechanical erosion of rocks in the sea.

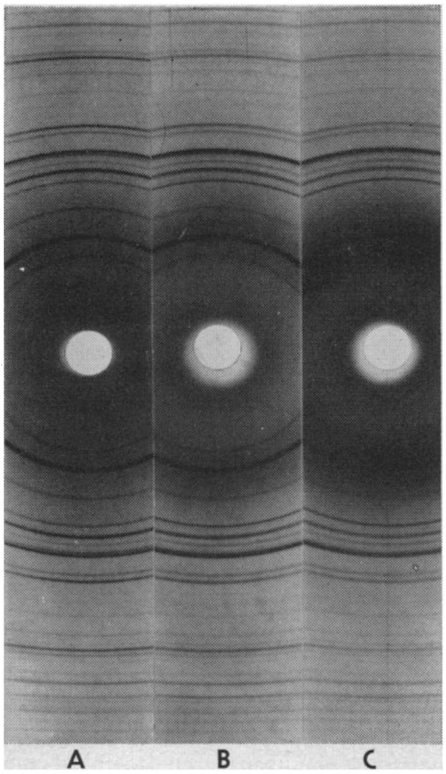

Fig. 1. X-ray diffraction photographs, Fe filtered Co radiation. *A*, reference goethite; *B*, single denticles of *Lottia gigantea*, Corona del Mar, California; *C*, single denticles of *Acmaea mitra*, Sitka, Alaska.

The teeth of the *Acmaea*, *Patella*, *Nomaeopelta*, and *Lottia* species investigated here were shown to be capped by goethite with a hardness close to 5. Hence these species are able to erode limestone and rocks with mineral grains in hardness close to that of their goethite denticles. The teeth of the species of *Fissurella*, *Diadora*, *Tegula*, and *Littorina* were shown to consist of organic compound with a hardness of less than 3. Therefore, these species are incapable of eroding limestone and most other rock types, except where mineral grains are already loosened by biochemical, physiochemical, or mechanical processes. These species must be grouped with the algal grazers. Large sediment volumes have been reported from the gut contents of species (*9*) which belong to this category. One source of the sediment particles has already been mentioned. Wave-transported sediment grains which are trapped by and agglutinated on the mucilagenous sheath of filamentous algae are another source of the ingested sediments.

More data are needed on the compounds which compose radular teeth in Mollusca. The sources of iron could be marine algae, which commonly enrich iron relative to sea water (*4*) and form a major part of the diet of chitons and the gastropods under consideration. The iron could also be derived from the dissolution of ingested sediment particles in their intestinal tract. It has been reported that hemocyanin is the oxygen-carrying transport pigment in the blood, and myoglobin occurs in the radular muscles of some of the chitons and gastropods under consideration (*10*). The localization of myoglobin in the radular muscles may have some bearing on the mechanisms of the precipitation of magnetite in the chiton denticles and of goethite in the denticles of the gastropods noted above. Clearly, there appears to be a fundamental biochemical problem involving iron-transport systems and the mechanisms for precipitation of goethite and magnetite by gastropods and chitons (*11, 12*).

H. A. Lowenstam
Division of Geological Sciences, California Institute of Technology, Pasadena

References and Notes

1. J. T. Tomlinson, *Veliger* **2**, 36 (1959).
2. H. A. Lowenstam, *Bull. Geol. Soc. Am.* **73**, 435 (1962).
3. J. Spek, *Z. Wiss. Zool.* **118**, 313 (1919).
4. A. P. Vinogradov, *The Elementary Chemical Composition of Marine Organisms* (Yale Univ. Press, New Haven, 1953).
5. E. J. Jones *et al.*, *J. Exptl. Biol.* **12**, 59 (1935).
6. To remove the organic fractions, samples of teeth from all species were treated with a Clorox solution (5.3 percent aqueous solution of sodium hypochlorite). Of the teeth from species of the genera *Acmaea*, *Patella*, *Nomaeopelta*, and *Lottia*, only the opaque denticles remained intact. The transparent teeth of the species from the other genera listed were entirely digested in the solution, indicating that they are composed of organic compounds.
7. R. N. Ginsburg, *Bull. Marine Sci. Gulf Caribbean* **3**, 55 (1953).
8. N. D. Newell, *Bull. Am. Museum Nat. Hist.* **109**, 311 (1956).
9. W. J. North, *Biol. Bull.* **106**, 185 (1954).
10. C. L. Prosser and F. A. Brown, Jr., *Comparative Animal Physiology* (Saunders, Philadelphia, ed. 2, 1961).
11. This study was supported by grants from Shell Development Co. and the Petroleum Research Fund of the Chemical Society.
12. This report is contribution No. 1085 from the Division of Geological Sciences, California Institute of Technology.

27 April 1962

Some Effects of Room Acoustics on Evoked Auditory Potentials

Abstract. Auditory potentials were recorded from bipolar electrodes chronically implanted in the cochlear nuclei of four cats. In a training box modified to reduce echoes these animals were exposed to clicks and tone pulses presented from an overhead speaker. Slight changes in the position of the animal in the resulting sound field produced marked changes in the potentials evoked from the cochlear nucleus. These phenomena were observed in the unanesthetized, unrestrained subjects as well as in those under Nembutal anesthesia. It is suggested that these acoustic effects complicate the analysis and interpretation of potentials evoked from the cochlear nucleus under conditions of habituation, shifts in attention, and learning.

When pulses of sound are introduced into a room or chamber a complex acoustic field is produced. Points of high and low sound pressure level result from the interaction of direct

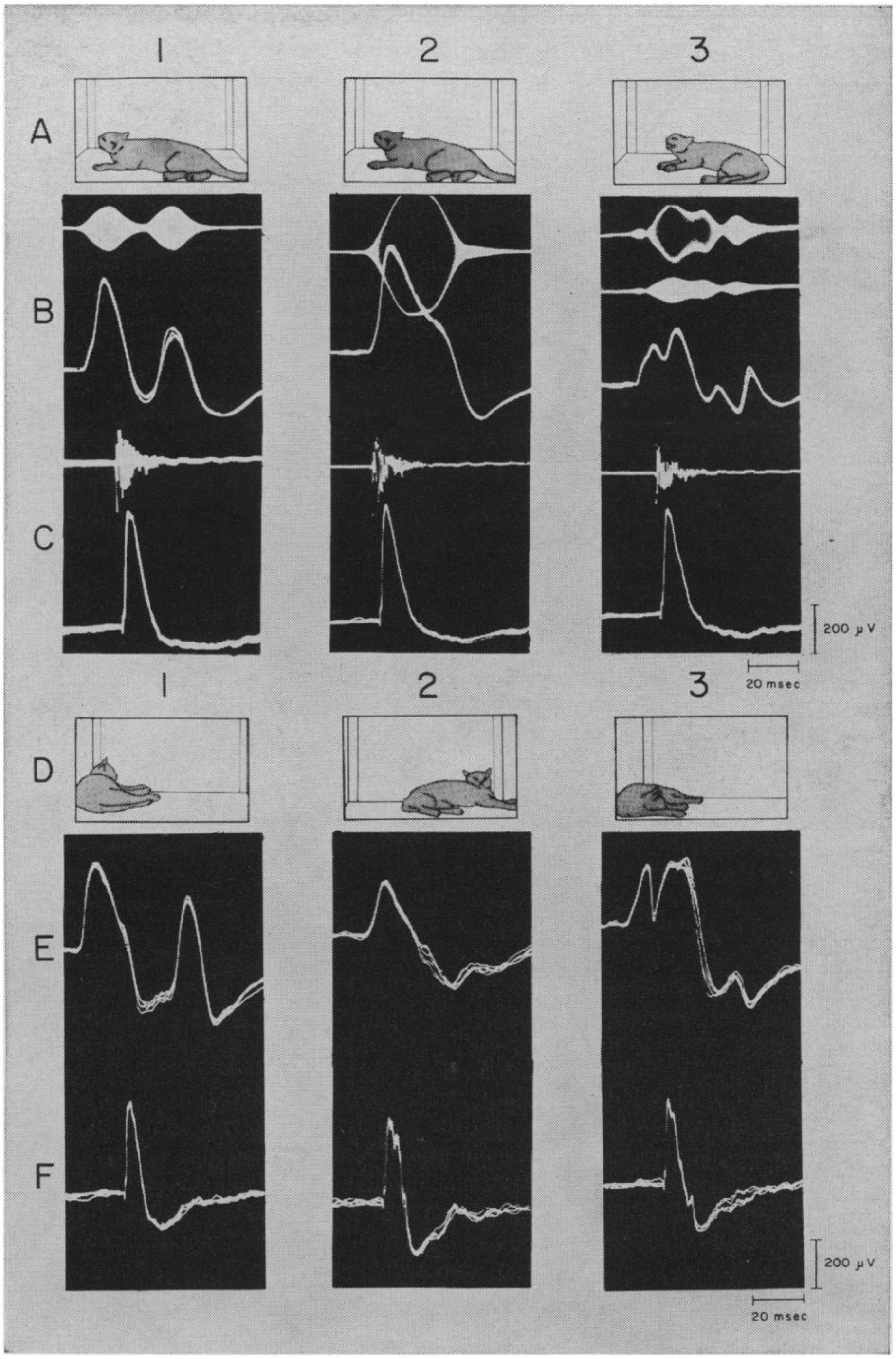

Fig. 1 (right). Influence on evoked auditory potentials (cochlear nucleus) of changes of position within an acoustic field. Top half of the figure (*ABC*) is from the anesthetized cat (Nembutal, 35 mg/kg). Bottom half (*DEF*) is from the same subject unanesthetized and unrestrained. For each condition data from three different positions are presented. (1, 2, 3). The anesthetized cat was moved ½ inch between *A*1 and *A*2. *A*3 is 3 inches from *A*1 with a slight change in head orientation. Acoustic stimuli were delivered at the rate of 1 per second from an overhead speaker. For *B* and *E*, there were tone pulses, 30 msec in duration, 3000 cy/sec. Clicks (1 msec square wave) were used at *C* and *F*. The top traces in *B* and *C* are from a microphone at the right ear. The bottom traces are bipolar recordings from the right cochlear nucleus. (No microphone trace is present at *E* and *F*.) In position *A*3 sound intensity is so reduced that an extra microphone trace at higher oscilloscope gain is added to reveal details of the sound-pulse envelope (*B*3).

and reverberant sound waves. The duration, amplitude, and envelope of the sound pulses (displayed oscilloscopically) change as the recording microphone is moved through the acoustic field. If microphone position is held constant, similar effects on the pulses are produced by changes in the intensity or frequency of the sound, or the location of the sound source. Changes in the acoustics of the chamber, including the presence of small objects such as a microphone, also influence the characteristics of the sound field. (*1*).

For several years the effect of these acoustic factors on evoked auditory potentials has been investigated in cats with chronically implanted bipolar electrodes. Results for the cochlear nucleus (CN) are reported here (*2*).

The experimental subjects were four adult cats with bipolar electrodes chronically implanted in the left and right cochlear nucleus. Electrode placements were verified histologically. Two acoustic stimuli were used: (i) tone pulses of fixed frequency, having a duration of 30 msec, and (ii) clicks of 1-msec duration. These were delivered at a rate of 1 per second from a loudspeaker centered 3 feet above the floor of a training box containing the cat. The training box was 29 inches long, 20 inches wide, and 16 inches high. To reduce echoes, the observation wall was of screen rather than glass, and the other inside walls were lined with sound-absorbent material. Bipolar recording of the CN potentials was displayed oscilloscopically, and five superimposed sweeps were taken for each picture. Records were taken from each cat under two conditions. Under Nembutal anesthesia (35 mg/kg) the animal was placed in the training box with its head fixed in a head-holder, and with a microphone located ½ inch from the ear. For a given position of the cat the responses of the cochlear nucleus and the microphone to tones and then to clicks were recorded. Similar recordings were obtained after each of a series of small changes in the cat's position. Both rotational and linear movements of the head were used, usually on the order of 5 degrees or ½ inch. Stimulus intensity was adjusted to 45 db at the microphone in the first position, and this setting of the audio amplification system remained constant through each series of positions. In the second condition the cat was unanesthetized and unrestrained. Since it was not feasible to keep a microphone at the cat's ear, initial stimulus intensity was set at 45 db for a central reference point in the training box. Each time the cat assumed a new position, recordings were taken of potentials evoked in the cochlear nucleus by tones and clicks. Under both conditions each position of the cat was photographed, and schematic drawings were prepared from these pictures.

Figure 1 presents data obtained from cat 15 which are representative of our experimental results. Recordings taken under anesthesia are shown in the top half of the figure. Three different positions of the cat are represented in row *A*. Row *B* shows the responses of the microphone and the right cochlear nucleus to tone pulses in each of these positions. Row *C* shows the corresponding responses to clicks. The same format is used in the bottom half of the figure to present results from the unanesthetized animal.

It is evident in the figure that small changes in the cat's position produce marked modifications in the responses of microphone and cochlear nucleus to tone pulses. It is equally apparent that the responses of the cochlear nucleus to tones follow variations in the envelope and amplitude of the sound pulse. The inverse correlation between envelope-complexity and peak-to-peak amplitude of the sound pulses is preserved in the responses of the cochlear nucleus. For example, the cat was moved ½ inch between *A*1 and *A*2, which resulted in a shift from double to single responses of both the microphone and the cochlear nucleus, along with an increase of peak-to-peak amplitude in both.

When click stimuli are used (Fig. 1, rows *C* and *F*) there is less change in microphone and CN responses as position is varied. In this acoustically deadened box decrements of over 50 percent in tone-evoked CN responses were observed, whereas the maximum decrement with clicks was 37 percent (cat 17). In a training box with the usual glass window and plywood walls, decrements as large as 50 percent have also been observed in click-evoked CN responses. These effects occur regardless of whether the tympanic membrane and ossicular chain are intact or destroyed. They are observed equally well from electrode placements yielding large or small CN potentials.

A number of recent reports (*3*) have described amplitude changes in evoked CN potentials during habituation, shifts in attention, and learning. Central neural influences have been cited as factors mediating these changes. Our results demonstrate another factor, namely, the powerful effects on evoked CN potentials produced by small differences in position of the animal within the acoustic field. These effects on evoked auditory potentials pose methodological and theoretical problems for the investigation of auditory electrophysiology and behavior. Specifically, habituation, shifts in attention, and learning are associated with characteristic changes in position of the animal. The acoustic effects of these position changes have to be isolated from central influences on CN potentials. The possibility that particular changes in CN potentials may reflect acoustic or central neural influences, or both, complicates the interpretation of such phenomena.

James T. Marsh
F. G. Worden
L. Hicks

Department of Psychiatry and
Brain Research Institute,
University of California
School of Medicine, Los Angeles

References and Notes

1. G. von Békésy, *Experiments in Hearing* (McGraw-Hill, New York, 1960), p. 369; H. Van den Ende, *Henry Ford Hosp. Med. Bull.* **8**, 299 (1960).
2. Results based on research supported by NIMH grant M-3831.
3. R. Hernández-Peón and H. Scherrer, *Federation Proc.* **14**, 71 (1955); ——— and M. Jouvet, *Science* **123**, 331 (1956); R. Galambos, G. Sheatz, V. G. Vernier, *ibid.* **123**, 376 (1956); M. Jouvet and R. Hernández-Peón, *Electroencephalog. Clin. Neurophysiol., Suppl.* **6**, 39 (1957); R. Galambos, in *The Central Nervous System and Behavior, 1st Conference*, M. A. B. Brazier, Ed. (Josiah Macy, Jr. Foundation, Madison Printing Co., Madison, N.J., 1958), p. 375; J. T. Marsh, D. A. McCarthy, G. Sheatz, R. Galambos, *Electroencephalog. Clin. Neurophysiol.* **13**, 224 (1961).

12 February 1962

An Apparently New Lethal Virus Disease of Infant Mice

Abstract. A hitherto unreported disease of infant mice is described. The agent reproduces upon passage, is filterable, is confined mainly to the gastrointestinal tract in infant and adult mice, causes death in infants and inapparent infection in adults, and is sensitive to mild heat, ether, and sodium deoxycholate. Unique and characteristic morphologic changes occur in the epithelial cells of the intestine in infected infant mice. These consist in the development of multinucleated giant cells designated "balloon cells."

A disease accompanied by diarrhea and death was noted in infants of a colony of C57BL mice in 1959. The disease was at first attributed to flagellates that were found in the intestinal contents; but when these were lost upon passage, and when bacteria-free filtrates of intestines with contents were found to be infectious, it seemed likely that a virus disease was being dealt with. Because the agent kills infant mice with regularity, because the original material has to date been diluted at least 10^{-25} by passage of intestinal filtrates, and because the agent is confined, primarily, to the intestinal tract, it has been tentatively designated "lethal intestinal virus of infant mice" (LIVIM). No description suggestive of the disease in mice has been found in the literature.

As noted above, diarrhea may occur in LIVIM infection, and mice manifesting the disease in mild form may simulate animals suffering with epizootic diarrhea of infant mice (EDIM) (*1*). The similarity ends there, however; for, unlike the EDIM-infected mouse, the LIVIM-infected mouse does not nurse, loses weight rapidly, becomes lethargic, and dies after a short period of cyanosis.

Upon sacrifice and autopsy of sick and dying animals, it is seen that gross pathology is limited to the digestive tract. The stomach is shrunken and devoid of milk. Bile-tinged material may be present throughout the intestinal lumen; and portions of the small intestine, distended by a large volume of gas, are so thin-walled that they may rupture during life.

Microscopically, infected mice show remarkable and unique changes. Numerous swollen multinucleated cells are noted throughout the intestinal tract but are most frequent in the small intestine. These cells appear to be modified epithelial cells. Phosphotungstic acid staining indicates that the nuclei are not separated by cell membranes. Of further interest is the fact that the nuclei have never been observed in any stage of mitosis. For want of a better term, to distinguish them from other multinucleated giant cells, and because of their appearance (Figs. 1 and 2), the cells are referred to as "balloon cells." Early in the course of the disease, many balloon cells display a distinct cytoplasmic basophilia. Later, more of them manifest cytoplasmic eosinophilia with a few large inclusions that are also eosinophilic. Inclusions are not seen in nuclei of balloon cells nor in neighboring normal cells.

In the small intestine the villi decrease in size and number. The mechanism of the disappearance of the cen-