

about an hour a day for several days. A total of 1300 different monosyllabic content words were presented to each observer—100 words from each of 13 intervals. Within each experimental session, words were presented in a random order with respect to word-frequency, except that each interval had to be represented 12 times in each block of 156 trials. The observers were instructed to respond with a monosyllabic word.

The intervals were set up according to data obtained from the Lorge Magazine Count (4). There are approximately 6500 monosyllabic content words in this count; therefore, each interval permitted approximately 500 different response words. The median word-frequency of the intervals ranged from 1 to 736 (in the Lorge sample of 4.5 million word occurrences). The stimulus words were selected randomly except for the attempt to control word length.

Figure 1 shows the results in terms of Eq. 1. (The axes, however, are in terms of number rather than probability.) Figure 1 indicates that  $p_c(s,r)$  is a constant proportion ( $k$ ) of  $p(s,r)$ , or that the probability of correct response is independent of the interval of the stimulus word when the stimulus and response words agree in interval. For 0 and +10 db,  $k$  is approximately 0.63 and 0.94, respectively. Linear curves with zero intercept are drawn to indicate the excellent agreement between Eq. 1 and the data.

Figure 2 shows the results in terms of Eq. 4 (in terms of number). Figure 2 indicates that  $p_c(s,r)$  is a linear function of  $p(r)$ , as predicted from Eq. 4. Since  $p(s,r)$  differs from  $p_c(s,r)$  only by the multiplicative constant  $k$ , Fig. 2 also indicates that  $p(s,r)$  is a linear function of  $p(r)$ . Thus, the effect of response bias—the tendency for  $p(r)$  to increase with the median word-frequency of the interval—is to increase  $p(s,r)$ .

Figures 1 and 2 support the response bias explanation by implying that response bias is a necessary condition for the word-frequency effect. When  $p(r)$  increases with the median word-frequency of the interval—and this is what we mean by response bias— $p(s,r)$  also increases, and since  $p_c(s,r)$  is proportional to  $p(s,r)$ , the word-frequency effect is obtained. The result of controlling response bias is to eliminate the word-frequency effect, as can be seen from the fact that  $p_c(s,r)/p(s,r)$  is a constant (Fig. 1). Thus words of high word-frequency are not identified more accurately than words of low word-frequency when response bias is controlled.

Even though response bias accounts fully for the word-frequency effect, the

magnitude of the obtained effect is greater than our mathematical interpretation of response bias explanation (Eqs. 1 and 2) can account for. The slopes obtained in Fig. 2 are considerably greater than the predicted slopes ( $k/n$ ). Values of 0.31 and 0.08 were obtained for +10 and 0 db, respectively, while 0.07 and 0.05 were predicted. This means that an increase in  $p(r)$  results in too great an increase in  $p(s,r)$  and, therefore, too great an increase in  $p_c(s,r)$ . If Eqs. 1 and 2 are accepted as the appropriate interpretation of acoustical equivalence, the implication seems to be that, contrary to the contention of response bias explanation, words do carry some acoustical information about their interval (5).

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4. E. L. Thorndike and I. Lorge, *The Teacher's Word Book of 30,000 Words* (Teachers College, Columbia University, New York, 1944). Actually we took advantage of a revised listing by frequency, which was kindly lent to us by Davis Howes of Massachusetts Institute of Technology.
5. This is technical report No. AFCCDD TR-60-34 of the Air Force Command and Control Development Division. This research supports project 7684, communication in noise, of the Air Research and Development Command's program in human performance.

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#### Relation between the Inorganic Chemistry and Biochemistry of Bone Mineralization

**Abstract.** In vitro experiments with saliva resulted in precipitation of a mineral substance (dahlite or carbonate hydroxyapatite) which is comparable in composition and crystal structure to oral calculus. Similar mineral substances were produced from synthetic solutions containing sodium phosphate and calcium chloride (in addition to a buffer) in the presence of carbonic anhydrase and available carbon dioxide. It is concluded that the carbonate ion is essential to precipitation of bone mineral and that the principal biochemical catalyst in vivo is carbonic anhydrase. Bacteria are not essential to the precipitation, but they probably play a secondary role in connection with the formation of oral calculus, urinary calculus, and so forth.

On the basis of an understanding of the inorganic composition and crystal chemistry of the tooth and bone mineral (1), it was predicted (i) that the carbonate (or bicarbonate) ion is es-

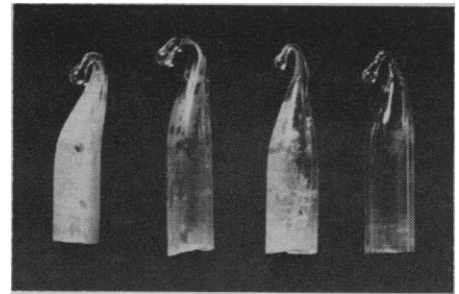


Fig. 1. Plummets immersed intermittently in (left to right): untreated whole saliva, saliva inactivated by heating, saliva to which carbonic anhydrase was added after heating, and saliva to which carbonic anhydrase and sulfanilamide were added after heating.

sential to the precipitation of the bone mineral, (ii) that the presence of a bacterial flora is not essential, and (iii) that some single biochemical substance, such as an enzyme, might catalyze the reaction and thereby govern whether or not mineralization takes place. The enzyme which immediately attracts attention is carbonic anhydrase.

Inasmuch as the mineral substance of the commonest type of oral calculus is dahlite (a carbonate hydroxyapatite)—and therefore essentially similar to bone and tooth mineral—it was decided to investigate in vitro those processes which are related to the formation of oral calculus.

Our first experiment consisted of repeatedly dipping small glass plummets into saliva by means of a motor-driven apparatus. The saliva was collected from persons who readily accumulated calculus. After 5 days an appreciable deposit appeared on the glass plummet which was intermittently immersed in untreated saliva (see Fig. 1). After the saliva had been boiled, the amount of solid deposited on the plummet was insignificant. However, if crystalline carbonic anhydrase was added to the saliva for which the enzyme had been inactivated by heating, again a heavy precipitate formed on the glass plummet. If, in addition to the enzyme, sulfanilamide was added to inhibit the activity of the enzyme (2), virtually no deposit was formed. During the course of these experiments the only source of carbon dioxide, other than what was initially present in the saliva and was not removed by heating, was laboratory air.

Two additional sets of experiments were particularly informative. Both of these were done with solutions containing sodium phosphate and calcium chloride. Although the solutions were not sterilized, no significant bacterial contamination could have occurred. First it was discovered that when the solutions were saturated with carbon

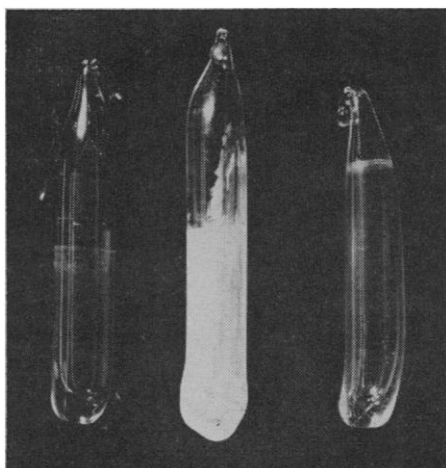


Fig. 2. Plummets immersed intermittently in synthetic solutions in an atmosphere of carbon dioxide (left to right): without enzyme, with enzyme, and with enzyme and inhibitor.

dioxide by bubbling the gas through them, the enzyme was not required in order for a deposit to form. These solutions presumably lost carbon dioxide until an optimum concentration of carbonate ions and pH favored precipitation.

However, when these synthetic solutions were not initially saturated with carbon dioxide and when the apparatus was enclosed in an atmosphere of carbon dioxide, the results (Fig. 2) were essentially similar to those obtained with boiled saliva: that is, no significant deposit was obtained unless carbonic anhydrase was added, and addition of sulfanilamide prevented formation of a deposit. These solutions contained also an organic buffer. The initial pH was 7.5 and no appreciable change took place accompanying the deposition.

In all cases the mineral depositions probably consisted of microcrystalline dahllite. By means of x-ray diffraction methods this fact was verified for three precipitates: the only deposit obtained from the second set of experiments with synthetic solutions (the one con-

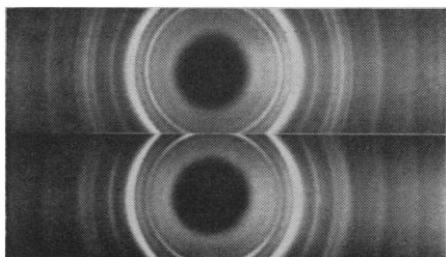


Fig. 3. Comparison of powder diffraction patterns of in vitro bone mineral precipitated from synthetic solution (top) and mineralized cartilage from the spinal column of a shark (bottom).

taining carbonic anhydrase) and the two significant deposits obtained with saliva (untreated saliva and boiled saliva to which carbonic anhydrase had been added). No evidence of the presence of a second crystalline phase was obtained (Fig. 3), but this does not necessarily prove that the nascent deposition has the crystal structure of an apatite. It merely proves that the stable inorganic substance in such a system is a carbonate hydroxyapatite (3), regardless of any inorganic precursor which might have existed (4).

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3. The carbonate content of an apatite cannot necessarily be demonstrated by x-ray diffraction methods for microcrystalline materials, but it was confirmed by liberation of bubbles of gas during dissolution of the deposits in hydrochloric acid.
4. The specimen of mineralized connective tissue of a shark, which we find to be crystallographically identical with bone, was supplied by Dr. Marshall R. Urist, Department of Surgery, University of California Medical Center, Los Angeles. The diffraction pattern indicates preferential orientation of the crystallites because the specimen was not powdered but was merely cut in the shape of a rod. This investigation was supported by a grant to the Ohio State University Research Foundation by the Procter & Gamble Co., Cincinnati.

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#### Relations between Whitefly and Sweetpotato Tissue in Transmission of Yellow Dwarf Virus

**Abstract.** One approach revealed the nature of the piercing-sucking-feeding mechanism of the abutilon whitefly, *Trialeurodes abutilonea*, a new vector of sweetpotato yellow dwarf virus. It showed the complete stylet from its origin in the rostrum to its termination in the plant's translocation stream. Another approach clearly delineated and confirmed the life cycle of the abutilon whitefly in relation to physical function and duration of each of the six stages.

Observations of sweetpotato plantings at Beltsville, Md., in 1955 and 1956, revealed relatively low percentages of natural spread of feathery mottle virosis. None of the leafhoppers in the same location were found to be vectors of the disease. In 1957 the abutilon whitefly, *Trialeurodes abutilonea* (Hald.), was found living on Indian Mallow, *Abutilon theophrasti* Medic. When an August drouth defoliated the weed, this insect was forced off the weed and onto the sweetpotato.

It was subsequently revealed to be a new virus vector (1).

Bennett (2) was probably the first to report on the tissue relations of a plant virus vector. By means of a camera-lucida drawing the stylet of the sugar beet leafhopper was shown in feeding position for transmission of curly-top virus. Fife and Frampton (3) demonstrated how the vector of curly-top utilizes the pH gradient for passing its stylet through the "acid" parenchyma into the "alkaline" phloem tissue, where it feeds.

The tobacco whitefly, *Bemisia tabaci* (Genn.), is known as a vector of several viruses in the subtropics. Pollard (4) studied its feeding habits in the Egyptian Sudan of Africa, where it spreads the cotton leaf curl virus and also causes other damage to cotton. In his anatomical studies a large majority of the insect stylets were observed penetrating to the depth of the phloem. His camera-lucida drawing showed the stylet in feeding position. It had perforated the epidermis and penetrated between the cells to the depth of the phloem. As pointed out by Avidov (5), this adult whitefly is well-qualified as a virus vector in Israel, where it is active about 5 weeks in the summer and 10 weeks in the winter. The same insect, *B. tabaci*, was also reported to be the vector of sweetpotato virus B in East Africa by Sheffield (6); subsequently, Girardeau (7) found that *B. tabaci* was the vector of a sweetpotato mottle mosaic in Georgia.

Concurrently, it was discovered (1, 8) that the whitefly, *Trialeurodes abutilonea* (Hald.), was the natural vector of sweetpotato yellow dwarf (feathery mottle) in Maryland; yellow dwarf is probably the same disease as was reported from Africa, Israel, and Georgia (6, 7, 9).

Direct micrurgical examination of the abutilon whitefly preceded the anatomical studies of the fixed material. The adult was the first and only stage of *T. abutilonea* observed in the field at Beltsville in late June each year since 1957. This means that pupae are the overwintering stage.

After the crawlers had been hatched, ordinarily they had stopped moving around and were feeding within an hour. After 2 days the rooted cuttings were ready for planting in peat pots for observation of the subsequent stages of development in the insect's life cycle. During 1958, five to ten larvae were transferred onto each of 50 lots of diseased cuttings and carried through to maturity with a percentage survival of 65. These transfer studies were repeated during 1959 and 1960, with essentially the same results.

A time interval of about 3 days was