

Letters

Carbonate in Apatites

In view of the authenticity usually attributed to the subjects discussed at symposia of the AAAS, it seems essential to comment on the chapter by A. S. Posner in *Calcification in Biological Systems* (1).

Posner's summary (1, p. 391) states: "Detailed data on the role of carbonate in hard tissue are included with the view expressed that this ion is admixed with the major apatite phase as a second finely divided calcite phase in the various carbonate-bearing apatites." Apparently the "detailed data" are those based on "preferential solubility" and the results obtained by several persons (1, p. 279) after heating various substances to various temperatures with variable results. The non sequiturs that are likely to arise from such experiments have been discussed by me (2).

Posner cites his investigations with several co-workers to show (i) that it has been possible to delicately refine the atomic parameters of "pure hydroxyapatite," and elsewhere (ii) that it is possible to prepare synthetic calcium-deficient apatite in which one of four symmetrically equivalent Ca atoms is missing. Both conclusions necessarily involve precise calculations based upon the x-ray diffraction intensity measurements, as well as definite knowledge that one is dealing with a completely homogeneous phase of accurately known composition. Posner *et al.* do not give complete analytical data on major constituents (CaO , P_2O_5 , and H_2O) for either set of experiments, and—far more important—they mention no tests to be certain that these precipitates were free from CO_2 . Their failure in the latter respect is particularly significant in view of the discovery by McConnell *et al.* (3) that the carbonate ion is essential to the formation of precipitates in systems approximating physiological compositions. Comparisons of the many measure-

ments of the unit cell dimensions of so-called hydroxyapatite (4) give rise to the question: Has pure (stoichiometric) hydroxyapatite ever been prepared? A corollary question is: How is it possible to obtain delicate refinement of the atomic parameters of something for which the composition is known solely by inference?

Far more important than these intricate details pertaining to "hydroxyapatite" are Posner's dismissals of contrary evidence and conclusions without any explanation. For example, he states (1, p. 380): "... there is no proof that the carbonate substitution takes place in scawtite." McConnell and Murdoch (5), on the other hand, supplied data from which they arrived at conclusions which are quite the opposite of those of Posner.

Elsewhere (1, p. 379) Posner says that x-ray diffraction intensity differences reported between fluorapatite and francolite (6) have been shown by him (7) and by Carlström (8) "to be due to orientation effects." Carlström, actually, was quoting Hendricks (9), who states (p. 186): "*In my opinion*, the principal reason that the sample of francolite gave this particular pattern was due to crystal orientation." (Italics added.) However, Hendricks' opinion does not coincide with the experimental data. For the (*hk*0) family of atomic planes, intensity measurements (automatically recorded with the North American Philips apparatus) show that although most of the prismatic reflections are of greater intensity for francolite, this is not true for all of them. In the absence of any theoretical explanation of how the intensities could increase for certain (*hk*0) reflections without doing so for all of them, it must be concluded that these intensity differences are *not* caused solely by orientation effects but must be caused by compositional differences. Furthermore, it cannot be shown that the differences in the fundamental periodicities are in any way related to

orientation effects, so there must be compositional differences which are capable of producing intensity differences, whether these intensity differences are measurable or not. My measurements definitely show intensity differences which cannot be attributed to preferential orientation. Several persons have confirmed the differences in fundamental periodicities.

Numerous other minor matters are discussed by Posner, but a thorough denial of most of his arguments surely exceeds the scope of this communication. Elsewhere (2) a series of data and conclusions are presented which lead to general conclusions that are diametrically opposed to those of Posner. The present communication is intended to consider merely some of Posner's arguments which have not been adequately considered elsewhere, but which are, nevertheless, incompatible with both the experimental data and accepted theory.

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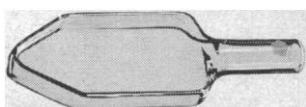
I welcome the opportunity to answer Duncan McConnell's comments on my review article entitled "The nature of the inorganic phase in calcified tissue" (1). McConnell objects to the concepts presented on carbonate-containing apatites and on the structure of various hydroxyapatites. His arguments will be dealt with in sequence.

The section of my article dealing with the carbonate problem was introduced with the statement, "The role of carbonate found in mineral tissue is still in debate." This treatment included references to both major viewpoints, (i) that the carbonate is substituted in the structure of apatite, and (ii) that carbonate is adsorbed, or ad-

ONE SOURCE

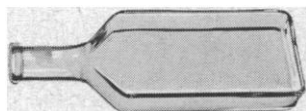
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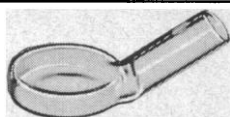
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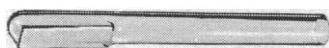
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mixed as a separate phase. In my conclusion I expressed the "view," that is, the opinion, that the bulk of the data support the exclusion of carbonate as a structural constituent in apatite. I made it clear that further evidence must be collected before this viewpoint can be held to be incontrovertible.

The article by McConnell and Murdoch (2) does not contain crystallographic proof that the carbonate is a structural constituent of scawtite. Without a detailed x-ray diffraction structure analysis, or some equivalent quantitative method which can assign, with certainty, position parameters to the constituent atoms, any structure must remain in doubt. Even if it were proved that carbonate appears as a structural constituent of this silicate mineral (scawtite), this would not constitute proof that the same situation holds for the basic calcium phosphates, that is, the apatites.

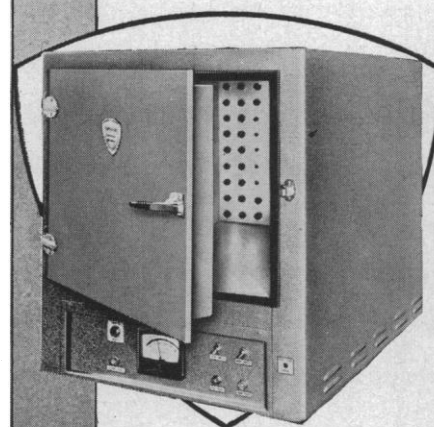
Hendricks (3), Carlström (4), and I (5) have stated that the x-ray diffraction patterns of francolite and fluorapatite as shown by McConnell (6) are different in relative intensity values, owing to an orientation effect. McConnell kindly supplied me with some of the francolite used in his study (6). It was possible to produce an oriented powder diffraction pattern such as he shows, or, if proper precautions were taken to avoid orientation, it was possible to produce a pattern coincident with fluorapatite. This result was reported to McConnell in personal conversation before he submitted his paper (6) for publication.

It is not true that the x-ray diffraction studies on apatite discussed by McConnell were performed on materials which were not analyzed. The single-crystal, x-ray diffraction refinement of the structure of hydroxyapatite was performed on well characterized crystals prepared in our laboratory (7). In addition, each sample of the calcium-deficient hydroxyapatites was shown to be a single phase and was characterized by chemical and physical analyses (8). Reference to the original papers will corroborate this statement.

Whether or not carbonate must be present for physiological mineralization is not a subject treated in my chapter (1) and needs no further discussion here. It is possible, by taking special precautions, to prepare hydroxy-

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apatite free of all carbonate content. Thus the presence of carbonate is not needed for the synthesis of hydroxyapatite.

The study of the exact nature of mineral tissue is fascinating work. There is room in this field for many viewpoints and dissenting theories. I think it is important to state clearly what is known and what is not known about these systems and I have attempted to do this in my chapter (1). I urge all workers in this field not to be satisfied with the half-answers in our possession now. We need more experiments, more facts, to define the atomic structure of hard tissue.

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References

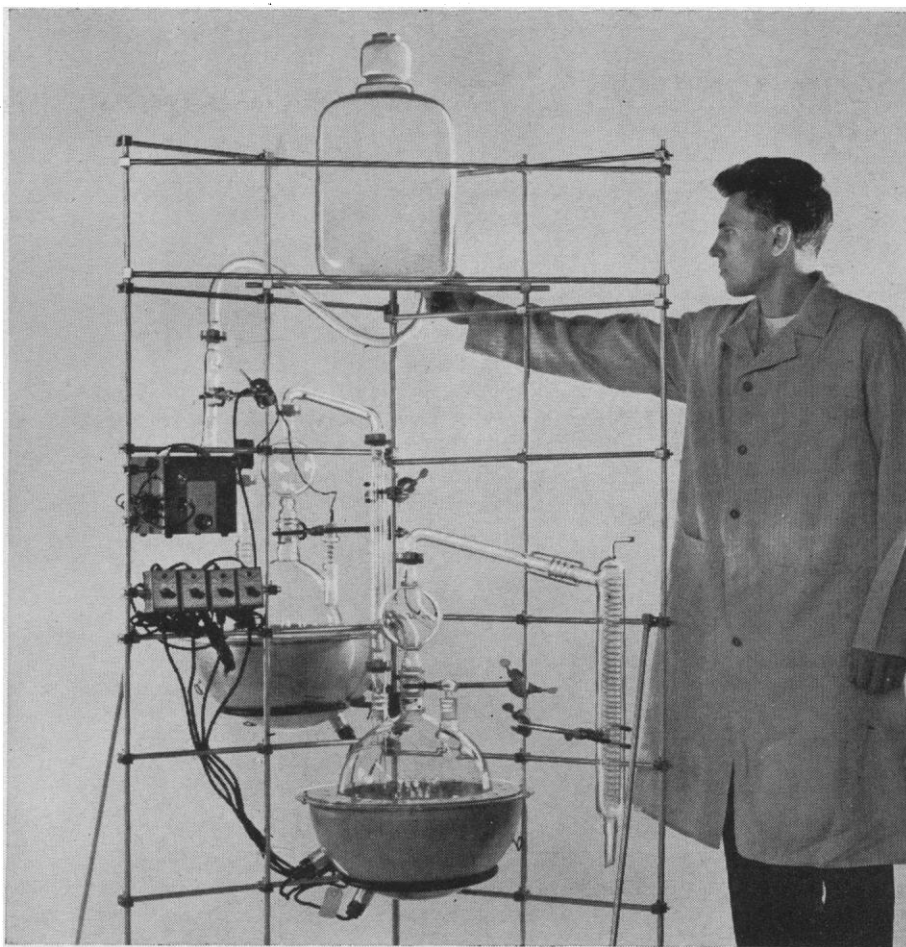
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Grants and Applicants

The game of measuring past scientists against present grant-reviewing policies [*Science* 133, 1040 (1961)] can be played without end. Freud was never psychoanalyzed, Mendel lacked training in genetics, Boas's degrees were not in anthropology, and Faraday's formal qualifications were belittled in his own time.

But these pioneers are not the applicants for today's research dollars. The request for a \$20,000 analyzer rarely comes from an amateur in science, and the retired school teacher does not seek \$50,000 (plus overhead) for studies on carcinogens. Current applicants make a point of their education, degrees, training, publications (and imagination) and expect to be judged accordingly.

It is true that a polished research plan does not automatically guarantee results. But no one has suggested that totally unplanned excursions are more productive. There are discovery-prone investigators and discovery-proof workers, and it is reasonable to bet on the former.



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