the complex because of endosmotic flow. The profound change in the mobility of the isotope when lactose is present is indicative of the formation of a complex.

The crystalline compound, $CaCl_2 \cdot$ lactose $\cdot 7H_2O$, was isolated and identified by Herrington (*7*), but no data were reported on the nature of the binding of Ca^{++} to the sugar, or on the characteristics of chelation. Our electrophoresis experiments conclusively demonstrate that the Ca^{++} forms a soluble, uncharged complex, similar to that observed for iron (*4*), but different from that of Herrington. Mills (*8*), in a series of paper-electrophoresis and chromatographic studies, showed that Ca^{++} and other divalent metals alter the migration patterns of polyols and suggested that complexes were formed. Wasserman and Lengemann (*3*) were not able to effect the solution of $CaHPO_4$ with lactose because of the unfavorable rate of the reverse reaction and the relationship between the stability constant of the Ca-lactose complex and the solubility product of the insoluble salt.

Several aspects of the regulation and absorption of Ca^{++} and other divalent metal ions, as influenced by sugars, are important to emphasize. There must be a high ratio of sugar to metal ion in order to have effective complex formation (*4*).

The rapidity of absorption of the sugar is extremely important. For this reason, it is difficult to correlate the stability of a Ca-sugar complex in vitro with its effectiveness in vivo, particularly where the literature indicates little agreement (*9*), and where quite different rates of intestinal transport for each particular sugar have been observed.

Our evidence for chelation of Ca^{++} by sugars offers a chemical explanation for some previous results of nutrition studies. Whether the complex is transported through the biological membranes or merely serves as a metal-ion buffer to maintain Ca^{++} in a form readily available for transport is yet to be determined (*10*).

PHILIP CHARLEY*
PAUL SALTMAN †

Department of Biochemistry,
University of Southern California,
Los Angeles 7

References and Notes

1. J. T. Irving, *Calcium Metabolism* (Methuen, London, 1957).
2. P. Fournier, Y. Dupuis, M. Susbrelle, A. Bourdeau, *Compt. Rend.* **148**, 265 (1959);
3. F. W. Lengemann, R. H. Wasserman, C. L. Comar, *J. Nutr.* **68**, 443 (1959).
3. F. W. Lengemann, *J. Nutr.* **69**, 23 (1959); R. H. Wasserman and F. W. Lengemann, *ibid.* **70**, 377 (1960).
4. P. Charley, C. Stitt, B. Sarkar, P. Saltman, *Biochim. Biophys. Acta* **69**, 313 (1963).
5. C. Stitt, P. J. Charley, E. M. Butt, P. Saltman, *Proc. Soc. Exptl. Biol. Med.* **110**, 70 (1962).
6. C. Long, in *Biochemists Handbook* (Spon, London, 1961), pp. 914–916.
7. B. L. Herrington, *J. Dairy Sci.* **17**, 805 (1934).
8. J. A. Mills, *Biochem. Biophys. Res. Commun.* **6**, 418 (1961).
9. Compare, for example, O. W. Vaughan and L. J. Filer, Jr., *J. Nutr.* **71**, 10 (1960) with R. H. Wasserman and C. L. Comar, *Proc. Soc. Exptl. Biol. Med.* **101**, 314 (1959). Note, however, that the former authors injected the Ca^{++} and sugar directly into the ileum, while the latter administered it *per os*.
10. Presented, in preliminary form, at the Conference on the transfer of calcium and strontium across biological membranes, Ithaca, N.Y., May 1962. Support from the U.S. Atomic Energy Commission and National Institutes of Health (AM-0548). We thank Clyde Stitt for valuable technical assistance and Arvan Fluharty for critical review.

* Present address: Truesdail Laboratories, Los Angeles, Calif.

† Research career development awardee, U.S. Public Health Service.

21 December 1962 ■

Calcium Carbonate: Factors Affecting Saturation in Ocean Waters off Bermuda

Abstract. *Bermuda marine waters are not in equilibrium with the bulk carbonate sediments with which they are in contact, and they are supersaturated with respect to pure calcite. This apparent supersaturation seems to result from a metastable equilibrium between sea water and the most soluble solid phase available locally in excess.*

The near-shore regions of Bermuda offer a wide variety of environments in which marine waters of differing temperatures and chemical compositions are in contact with relatively pure carbonate sediments. The island thus offers an ideal setting in which to study carbonate sediment–sea-water systems. We have made a preliminary saturometer-type investigation (*1*) to establish whether the waters are in equilibrium with the subjacent sediment. The results were unexpected.

Samples of sea water and bottom sediment were collected by skin-diving and bottom-sampler techniques from five localities: an open coast environment at Tobacco Bay on the northeast end of the island; a shallow semi-restricted environment in Ferry Reach near the Bermuda Biological Station; and from three locations at depths of 3, 15, and 78 feet in Harrington Sound, an enclosed lagoon. The *p*H of each water sample was measured immediately upon collection (*p*H, Table 1). An excess of a solid carbonate phase was then added, and a glass electrode was inserted into the sediment. The indicated *p*H was then recorded at 5-minute intervals until no further change could be observed (generally about 30 minutes). The difference between the observed final *p*H in the presence of excess solid and the original *p*H of the water sample was then recorded (ΔpH, Table 1). All measurements were made with a Beckman model G *p*H meter, which has a nominal precision of ± 0.01 *p*H unit and an accuracy of ± 0.02 *p*H unit.

The interpretation of the data presented in Table 1 is not unambiguous. Using pure calcium carbonate as the solid phase and solutions of both distilled water and artificial sea water, Weyl (*1*) has shown that saturometer measurements made as described will show positive values of ΔpH when the solution is initially undersaturated, negative values when the solution is initially oversaturated, and no change when the solution is in equilibrium with the added solid phase. Weyl's results show further that the magnitude of the observed *p*H change will be proportional to the degree of over- or undersaturation. When the solid phase used is a solid solution of variable composition, however, and the fluid is a phase as complex as sea water, the interpretation becomes less straightforward. Differential complexing of the carbonate-mineral cations by *p*H-sensitive ions in sea water or peritectic exchange of the solid solution cations may occasion *p*H changes not directly related to the saturation state of the water sample. Such changes would be small, however, and in view of the large *p*H changes observed in the present study the data in Table 1 are interpreted, qualitatively, as though they relate to a simple binary system.

So interpreted, the data in Table 1 appear to indicate that only the deepest water sample from Harrington Sound is in equilibrium with the subjacent bulk sediment. Even this sample is in equilibrium only with a 3-inch surface layer of ooze containing more than 90 percent aragonite. All of the remaining samples appear to be oversaturated with respect to the sediment with which they

Table 1. Saturometer results expressed as change in observed pH, for five water samples from Bermuda. Positive values of ΔpH are interpreted as indicating undersaturation; negative values, supersaturation. The solid phases used are fully identified in the text.

Depth (ft)	Initial pH	Observed pH change with respect to:				
		Subjacent sediment	Pure and low-Mg calcite	Aragonite and med-.Mg calcite	High-Mg calcite	
					Coarse	Fine
			Tobacco Bay (after rain)			
	8.22	−0.28				
			Ferry Reach			
	8.14		−0.52	−0.22	−0.46*	+0.17
			Harrington Sound			
3	8.23	−0.40	−0.50	−0.25	−0.22	−0.07
15	8.22	−0.46				−0.03
78	7.44	−0.01 −0.46†				+0.21

* This reading appears to be anomalous. † Determination made with light grey sediment from below the 3-inch layer of surface ooze. The surface ooze is a dark grey, organic-rich mud which evolves H_2S when disturbed.

are in contact. This observation poses the problem of determining the mechanism controlling the activities of dissolved carbonate species.

Bermuda sediments are made up of a variety of skeletal fragments. These are composed not only of calcite but also of aragonite and magnesian calcite solid solutions of a wide range of compositions (*2*). The solubility of aragonite is substantially greater than that of calcite, and the solubility of magnesian calcites increases with increased magnesium content (*3*). For this reason, saturometer experiments were repeated on several of the same water samples using single-phase carbonates of various compositions in an attempt to determine whether the solutions were in equilibrium with one of the more soluble phases. The single-phase carbonate minerals used, with the exception of reagent-grade calcium carbonate (calcite), were derived from the skeletons of organisms common in Bermuda waters: aragonite from *Oculina*, a coral; slightly magnesian calcite (less than 2 percent $MgCO_3$) from *Pinna*, a pelecypod; moderately magnesian calcite (9.5 percent $MgCO_3$) from *Plexaura*, an alcyonarian; and highly magnesian calcite (18 percent $MgCO_3$) from *Amphiroa*, a red alga.

When crushed samples (approximate diameter, 10 μ) of these carbonates were used in the saturometer, it was found that the Harrington Sound and Ferry Reach waters were greatly oversaturated with respect to calcite and slightly magnesian calcite, and somewhat less oversaturated with respect to aragonite and moderately magnesian calcite. The moderately magnesian calcite used in the experiments gave results almost identical with pure aragonite, as would be expected from the relative solubilities reported by Chave *et al.* (*3*). The water samples were most nearly in equilibrium with the highly magnesian calcite, though all appeared to be somewhat oversaturated even with respect to this relatively very soluble phase.

The solubility of carbonate minerals can be substantially increased by grinding (*4*). This effect results in part from the relatively high surface energies of the smaller particles (*5*) and in part from the energy contributed by lattice distortions introduced by grinding. When finely ground (particle diameter less than 1 μ) highly magnesian calcite (*Amphiroa*) was placed in the saturometer, the results indicated that the waters from depths of 3 and 15 feet in Harrington Sound were nearly in equilibrium with the solid phase, while the water samples from Ferry Reach and the 78-foot depth in Harrington Sound were undersaturated. The water samples from the wave zone, where constant stirring and grinding of the sediment should produce an abundance of extremely fine particles, appear to be in equilibrium with carbonate material even more soluble than the finely ground highly magnesian calcite employed in the experiment, while water samples from quieter areas appear to be in equilibrium with some slightly less soluble phase.

The saturometer experiments thus indicate that all of the water samples examined were in equilibrium with solid phases substantially more soluble than pure calcite, and that the amount of dissolved carbonate present in each sample is related to the nature of the environment from which the sample was collected.

It must be emphasized that these measurements do not provide direct evidence of the mechanism responsible in nature for the saturation state of the water samples. The saturometer shows only whether the activity of bicarbonate ion (the dominant species) in the sample is greater than, less than, or equal to that which would obtain in a saturated solution of the particular solid phase used in the saturometer. In the present case, however, two factors strongly suggest that the saturation state of the samples is controlled in the natural environment by a combination of grain size and the magnesium content of the carbonate phases present in the sediment. First, the amount of dissolved carbonate in each sample appears to bear a direct relationship to the character of the natural environment, and second, the subjacent sediment in each case includes an excess of that solid phase with which the solution may be shown to be in equilibrium in the saturometer.

We may thus account for the apparent supersaturation of the Bermuda water samples with respect to pure calcite by inferring that the saturation state of each sample is controlled by the solubility of the solid phases locally available. This conclusion suggests that the activity of dissolved carbonate species in Bermuda waters may be determined by a metastable equilibrium between sea water and the most soluble solid phase present in the sediment. Whatever the local controlling phase, the more soluble carbonate minerals, particularly in the finer size ranges, should be progressively removed by solution until only the most stable phase, coarse calcite, remains. The conclusion thus provides a mechanism to account for the observations reported by Chave (*2*) (*6*).

ROBERT F. SCHMALZ
Pennsylvania State University,
University Park

KEITH E. CHAVE
Marine Science Center, Lehigh
University, Bethlehem, Pennsylvania

References and Notes

1. P. K. Weyl, *J. Geol.* **69**, 32 (1961).
2. K. E. Chave, *Limnol. Oceanogr.* **7**, 218 (1962).
3. ———, K. S. Deffeyes, P. K. Weyl, R. M. Garrels, M. E. Thompson, *Science* **137**, 33 (1962).
4. R. M. Garrels, M. E. Thompson, R. Seiver, *Am. J. Sci.* **258**, 402 (1960).
5. R. F. Schmalz, in preparation.
6. Contribution No. 326 from the Bermuda Biological Station: contribution No. 63-1 from the Marine Science Center, Lehigh University; contribution No. 62-27 from the Mineral Industries Experiment Station, Pennsylvania State University. Supported by grants from the Petroleum Research Fund of the American Chemical Society.

16 January 1963