lae which had not been expelled. It is not known whether there was reinfection of the bleached corals by motile zoospores such as those described by Freudenthal (*7*) for *Symbiodinium microadriaticum*. Even in those colonies that lost only part of their zooxanthellae, as shown by their light yellowish-brown color, return to normal took between 6 and 10 weeks; if turnover rates are of the order postulated by Odum and Odum (*8*) regeneration of the zooxanthellae should have occurred in less than 2 weeks.

The effect of wholesale depletion of zooxanthellae upon a reef community is difficult to evaluate, for the functional relationship of these algae to their coelenterate hosts differs from group to group and in most cases is still imperfectly understood (*9*). Among the reef-building Milleporidae and Scleractinia, all of which are carnivores specialized for feeding on plankton (*10*), the most important consequence of the loss of zooxanthellae would be a general lowering of the skeletal calcification rate in the affected zones of the reef. It has been shown (*11*) that the calcium deposition rate of reef corals is much greater in bright light than in darkness, and that this effect occurs only in the presence of zooxanthellar photosynthesis. In the absence of the zooxanthellae, the calcification rate of reef corals falls to very low levels and becomes independent of the light intensity (*12*).

The possible consequences of zooxanthella depletion on the Actiniaria (for example, *Stoichactis*) and the Zoanthidea (*Palythoa* and *Zoanthus*) are a matter for speculation since there is little information about the function of the zooxanthellae in these noncalcareous forms. No signs of starvation were noted in bleached *Stoichactis* and *Palythoa* even 4 to 6 weeks after they had lost their zooxanthellae. Indeed I often saw such individuals actively feeding, the former on small fish and pteropods, the latter on smaller zooplankters. Evidently, like the reef corals, these two forms do not depend on zooxanthellae for the bulk of their nutritional requirements. On the other hand, *Zonathus sociatus* was the only form that could retain most of its zooxanthellae even when exposed to an osmotic stress so severe as to kill all other reef biota; it was also the only species which I never observed to feed on zooplankton, or any other food offered to it. This evidence supports the suggestion that *Z. sociatus* may be nutritionally dependent on the photosynthetic products of its zooxanthellae.

The most probable cause of the large-scale bleaching in the Port Royal reefs was contact with low-salinity surface water, but as yet nothing is known of the concentration range necessary to produce expulsion of zooxanthellae from millepores, corals, zoanthids and actinians. According to Wells (*13*) the optimum salinity for reef corals is about 34 to 36 per mil; salinities as low as 27 per mil are tolerated, but influxes of fresh water for even short periods of time are said to be fatal. The extreme rapidity with which zooxanthellae were extruded in the inundated parts of the reef suggests that exposure to reduced salinity under controlled conditions may constitute a practical method for speedy production of healthy bleached colonies for experimental purposes.

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References and Notes

1. C. M. Yonge and A. G. Nicholls, *Sci. Rept. Brit. Great Barrier Reef Expedition* **1**, No. 6, 135 (1931).
2. ———, *ibid.* **1**, No. 7, 177 (1931).
3. D. F. Squires, *Nature* **195**, 361 (1962).
4. I. M. Goodbody, *Ecology* **42**, 150 (1961).
5. S. M. Marshall and A. P. Orr, *Sci. Rept. Brit. Great Barrier Reef Expedition* **1**, No. 5, 93 (1931).
6. J. Verwey, *Pacific Sci. Cong. 4th* **2A**, 277 (1929).
7. H. D. Freudenthal, *J. Protozool.* **9**, 45 (1962).
8. H. T. Odum and E. P. Odum, *Ecol. Monogr.* **25**, 291 (1955).
9. M. Droop, *Symp. Soc. Gen. Microbiol.* **13**, 172 (1963).
10. C. M. Yonge, *Sci. Rept. Brit. Great Barrier Reef Expedition* **1**, No. 2, 13 (1930). Yonge's studies of the carnivorous feeding habits of Indo-Pacific reef corals have been confirmed by me on eight genera of West Indian reef-building corals and three non-reef-building species. Contrary to the accepted idea that reef corals are night feeders, I have found that most reef corals will feed in broad daylight if zooplankton is present in large amounts.
11. S. Kawaguti and D. Sakumoto, *Bull. Oceanogr. Inst. Taiwan* **4**, 65 (1948); T. F. Goreau, *Biol. Bull.* **116**, 59 (1959); ——— and N. I. Goreau, *ibid.* **117**, 239 (1959).
12. T. F. Goreau and N. I. Goreau, *Biol. Bull.* **118**, 419 (1960).
13. J. W. Wells, in "Treatise on marine ecology and paleoecology," J. W. Hedgpeth, Ed. *Geol. Soc. Am. Mem.* **67**, No. 1, 1087 (1957).
14. I thank Barry Wade for the salinity data in Table 1. This work was supported by NSF grant G-14589 and contract Nonr 2306(00) with the Office of Naval Research.

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Silica Source in Soil Solutions

Abstract. *Heat of solution of silica in soil saturation pastes was calculated from silica solubilities and agreed with values for solid silicic acid. The immediate source of silica in soil solutions apparently is solid silicic acid. The solution of silica from soils exhibited three stages of constant silica solubility.*

Silica and silicates form a major portion of the mineral matter of most soils. Although soil minerals have been extensively studied, confusion and uncertainty exists concerning the forms and properties of silica in soil. McKeague and Cline (*1*) have reviewed the extensive literature on silica in soils. They concluded that the silica in soil solutions exists almost entirely as monosilicic acid (H_4SiO_4). Furthermore the source of the silica was presumed to be a disorganized surface layer on quartz and perhaps other silicates. This study was undertaken to secure more definite information on the solubility of silica in soils.

Silica was determined colorimetrically in solutions extracted from saturated soil pastes prepared in a standard manner (*2*) with silica-free distilled water. Extraction was performed in a pressure extraction apparatus at 7.0 kg/cm^2 (gage) in a temperature-controlled water bath at 5°, 15°, 23° (room temperature), 40°, and 55°C. Samples were placed in the bath in sealed containers for 16 hours and then transferred to the extractor, which was placed in the water bath for at least 20 minutes to return the samples to the bath temperature.

Table 1. Heat of solution of silica computed from silica solubility in soil saturation pastes.

Soil type	Great soil group	Heat of solution (kcal/mole)
Unnamed loam*	Sierozem	4.2
Unnamed loam	Sierozem	4.2
Unnamed silt loam	Sierozem	4.0
Placeritos silt loam	Alluvial	4.2
Humboldt silty clay loam	Humic-Gley	4.2
Carson clay*	Humic-Gley	5.0
Carson clay	Humic-Gley	5.2

* Virgin site, all others irrigated fields. Samples represent the top 6 inches of the soil in all cases.

Plastic laboratory ware was used for all solutions except that the colorimeter tubes were glass. Silica standards were prepared by gravimetric analysis (see *2*).

In preliminary trials it was found that after 16 hours, silica content of soil saturation pastes had reached stable values. Silica solubilities (logarithm of molal silica solubility) at the five temperatures were plotted against the reciprocal of absolute temperature. Each value plotted was the average of triplicate determinations. Straight lines were obtained for each of seven soil samples. The slopes of the lines were determined by the method of least squares.

In Table 1 are presented the heats of solution of silica, calculated from the slopes of the curves. The three unnamed Sierozem soil samples, all from Nevada, were taken in a recently settled area north of Winnemucca. The Placeritos and Humboldt samples were from the flood plain and delta deposits, respectively, of the Humboldt River near Lovelock. The Carson series recently developed from shallow lake deposits near Fallon. The reason for the higher values of heat of solution of Carson samples is not immediately evident. It may be related to the origin of the parent material of the soil. Irrigation apparently has not modified the silica of either the unnamed loam or the Carson clay sufficiently to change heat of solution.

Heats of solution of silica in water have been calculated (*4*) on the basis of thermodynamic values for silica forms. The following heats of solution were reported: Amorphous silica: + 2.65 kcal/mole; quartz: + 7.34 kcal/mole. For solid silicic acid the heat of solution was computed as approximately 4.5 kcal/mole. The values obtained in this study are in good agreement with this heat of solution. Therefore it is highly probable that the immediate source of silica in saturation-paste extract of soil is monosilicic acid. As suggested by McKeague and Cline (*5*), the monosilicic acid is probably absorbed on the surfaces of soil silicates, including quartz.

An initial high solubility of silica is frequently noted in studies of the solution of silica from quartz and other forms of silica (*6*). This high solubility has been associated with an easily soluble layer that may be at least a part of the disorganized layer reported to be on the surface of quartz. An experiment was conducted to determine the extent of the easily soluble layer in soils. Duplicate 30-gram samples of Humboldt silty clay loam and Placeritos silt loam were placed in plastic bottles with 150 ml of silica-free water. The samples were shaken on a wrist-action shaker controlled by an automatic timer to run 1 minute out of each 10 minutes. After more than 48 hours at approximately 23°C (room temperature controlled by an air-conditioner), solid sodium chloride was added to the samples to cause flocculation of the clay, the suspension was cleared by centrifuging, 130-ml samples were removed, an equal volume of water was added, and the shaking was repeated. Silica was determined on the clear solution, and the quantity of extracted silica was calculated.

As shown in Fig. 1, the extraction of silica exhibited three readily separated stages in which the silica concentration of the solution was constant at the values noted in Fig. 1. Extrapolation to the ordinate of the straight lines formed by the data for the first stage of extraction gave an estimate of the easily soluble layer of 180 μg of silica per gram for the Placeritos samples and 150 μg/g for the Humboldt samples. Further extractions removed silica from the samples at lower yet constant rates, with mean solution concentrations as shown in Fig. 1. Extrapolation of the second and third slopes gave quite different estimates of the easily soluble layer, in agreement with the data of Bergman and Patterson (*7*), who found that their estimate of the easily soluble layer of quartz depended upon the du-

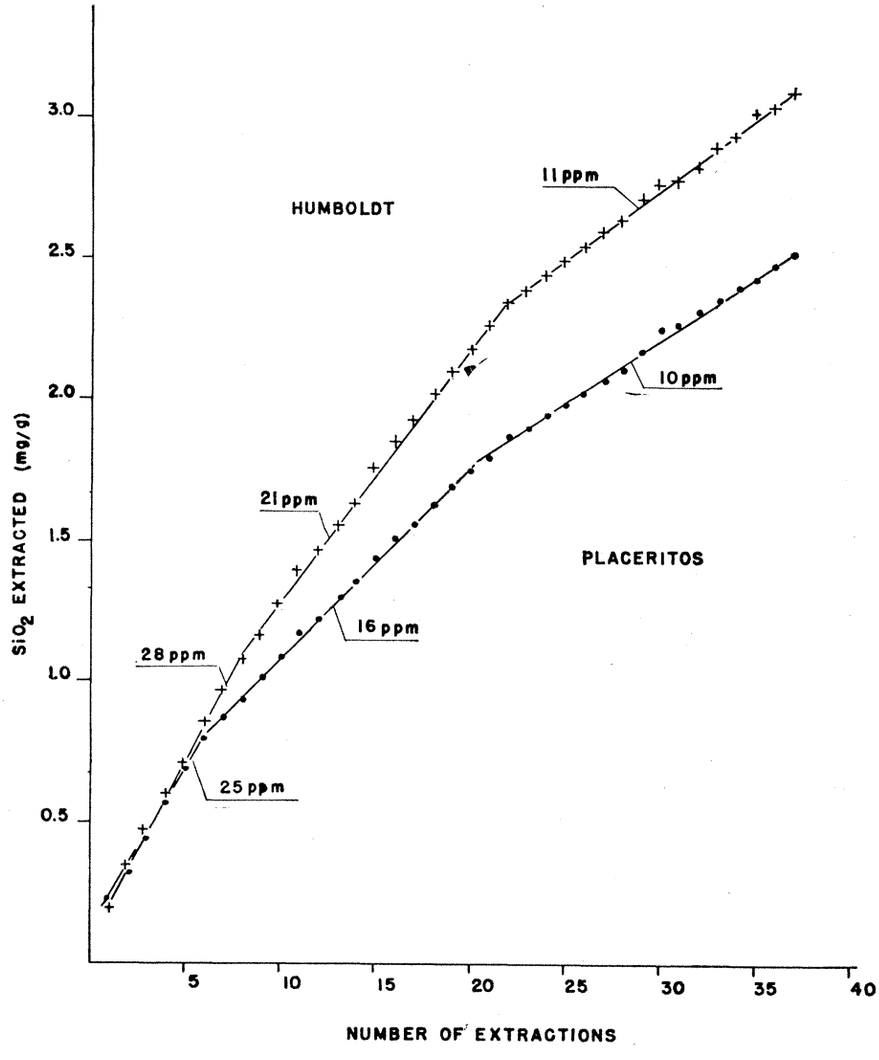

Fig. 1. Relation of number of 130-ml extractions to total SiO_2 extracted from 30-gram samples of two soils. The concentrations shown are the mean concentrations of the extracting solution calculated from the slopes of the straight lines.

ration of the extraction considered. The intercepts can be used to estimate quantities of forms of silica with different solubilities in the samples. Placeritos samples apparently contain 180 μg of a readily soluble form of silica per gram (H_4SiO_4), 220 μg/g of a second form of silica of lower solubility, and 500 μg/g of a third type. For the Humboldt silty clay loam, the corresponding figures are 150, 240, and 860 μg/g. In comparison, the saturation pastes used to determine the heat of solution showed 19 μg/g from Placeritos silt loam samples and 25 μg/g from the Humboldt silty clay loam samples. Approximately one-tenth of the solid silicic acid on the surface of the soil samples was sufficient to produce an equilibrium level of silica in the soil paste. At least one more slope would be anticipated in the extraction curve, since the solubility of quartz, approximately 6 parts per million, had not yet been reached. Under natural conditions of much slower extraction by downward moving water, equilibrium could be expected between silicic acid and the second and third types of silica postulated here as well as with quartz. The origin of the second and third types of silica is not clear: it could possibly be amorphous silica of plant origin or perhaps from the volcanic ash prevalent in northwestern Nevada.

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References and Notes

1. J. A. McKeague and M. G. Cline, *Advan. Agron.* **15**, 339 (1963).
2. L. A. Richards, Ed., *Diagnosis and Improvement of Saline and Alkali Soils, Agriculture Handbook No. 60* (U.S. Dept. Agriculture, 1954). "At saturation a soil paste glistens as it reflects light, flows slightly when the container is tipped, and the paste slides freely and cleanly off a spatula for all soils but those with a high clay content."
3. Baroid Division, National Lead Co.
4. S. A. Greenberg, *J. Phys. Chem.* **61**, 196 (1957).
5. J. A. McKeague and M. G. Cline, *Can. J. Soil Sci.* **43**, 83 (1963).
6. P. F. Holt and D. T. King, *J. Chem. Soc.* **1955**, 773 (1955).
7. I. Bergman and M. S. Paterson, *J. Appl. Chem.* **11**, 367 (1961).
8. This report is Nevada Agricultural Experiment Station Journal Series 9. This work was supported in part by Western Regional Research Project W-66.

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Accuracy of Bone Mineral Measurement

Abstract. *The use of nearly monochromatic radiation (I^{125}) for the direct determination of bone mineral absorption in cadaver materials indicates that underlying bone components can be measured accurately on the flesh-covered forearm. The correlation of the bone mass, determined by this scanning technique, with actual bone weight is 0.96. The method is the most accurate yet demonstrated for bone with overlying tissue, and may prove suitable for these studies in vivo where it has not been possible to use previous roentgenologic techniques.*

An improved method for measuring bone mineral in vivo, in which direct measurement is made of monochromatic photon absorption, was described by Cameron and co-workers (*1*). Theoretically, the use of monochromatic radiation should reduce the large uncertainties of the absorption coefficients associated with polychromatic x-ray beams used in other techniques. Use of a well-collimated radioisotope source and a collimated scintillation detector both reduces the effects of scattering and eliminates the film errors of roentgenographic densitometry. The results of preliminary investigations with Cameron's technique have indicated that the theoretical expectations are justified and that accurate estimates of bone mineral content may be made on flesh-covered extremities.

A number of other techniques for determining bone mineral content in vivo are now being used in laboratories here and in other countries. The most common are those based on transmission of polychromatic x-radiation as estimated by film densitometry. Several such methods were developed at the Pennsylvania State University (*2*), and have been shown to be accurate for excised long bones, but when bones with greater overlying tissue than the phalanges are used a substantial error is introduced (*3*, *4*). This error is not eliminated by either statistical or experimental control, and is apparently due to the polychromatic radiation, scattering, and film errors. The lack of accuracy restricts the use of the older densitometric methods for studies in vivo which might be of nutritional, gerontological, or clinical interest (*3*, *5*).

In the method developed by Cameron (*1*), a collimated I^{125} photon source (27.3 kev) is passed across a forearm and the transmission measured with a collimated scintillation crystal, pulse height analyzer, and scaler. Transmission counts are recorded from the scaler for 7-second periods at each 1-mm interval across the forearm. Logarithms of these counts are plotted at equal intervals on semi-logarithmic graph paper. The two smooth curves representing tissue absorption and bone absorption are drawn through the plotted points, and the area between the two curves is measured by planimeter. The scan area determined in this manner is theoretically proportional to the cross-sectional bone absorption and hence proportional to the mass of bone mineral in the line traversed during the scan.

We have determined the areas for 35 scans across the radius and ulna on two cadavers. Form-fitting pieces of tissue-equivalent material (Mix-D), of the same composition as previously described (*1*), were used as a bolus to maintain equal "tissue" thickness across the forearm. Overlying flesh was removed after the scans were made, and at each of the scan locations 1-cm sections were accurately cut from the bone on a milling machine. These sections were defatted in acetone for over 48 hours, dried to constant weight, and the dry fat-free weight was determined. The sections were then ashed to constant weight (600°C for 12 hours or 400°C for 24 hours) and the weight of the ash obtained.

The dry fat-free weight and the ash weight are shown in Fig. 1 plotted

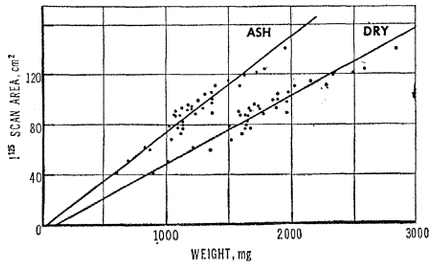

Fig. 1. The I^{125} scan area on the flesh-covered forearm plotted against the ash weight (ASH) and the dry fat-free weight (DRY) of 1-cm sections of underlying bone ($n = 35$).