tyrosine structure has five residues unpaired. The transition to the left-hand limb is made in both cases by DiMeG. The transition from the left to the upper limb is made in an identical fashion in both structures—by two unbonded nucleotides.

In addition to the DiMeG and T-ψ-C-G residues being in homologous positions, it is apparent from the cloverleaf arrangements that ψ is found only in the lower and right-hand loops, while DiHU is found in the left-hand loop and in the transition from the right-hand to the lower limb.

The alanine tRNA has no unusual nucleotides in the regions that are proposed to be hydrogen-bonded. The tyrosine RNA, however, has 2MeG and ψ in locations that are presumably hydrogen-bonded.

It might be significant, on the other hand, that bases that presumably cannot form a hydrogen-bonded base pair such as DiMeG, DiMeA, 1MeI, and 1MeG, are all located in regions depicted as being not hydrogen-bonded.

In both structures only the G- in the bottom loop is cleaved by ribonuclease T1 at 0°C and 0.02*M* $MgCl_2$. It is difficult to understand this specificity unless the other loops are rendered inaccessible. It is possible that the two lateral loops can form hydrogen bonds with each other. In both cases, the bases within the arrows could form two GC, two GU, and one or two AU type base pairs depending on whether the MeA in the right-hand loop of the tyrosine tRNA can form a base pair. The above also assumes that DiHU can form effective hydrogen bonds.

The activating enzyme recognition site is still not obvious. In addition to the anticodon (*14*), the DiHU containing left-hand limb and the unbonded residues between the lower and right-hand limbs might be likely candidates.

The sequences G-ψ-A in the lower loop and G-U-A between the left and upper limbs are the only sequences that satisfy all the requirements for the anticodon for tyrosine (*15*), since there is no A-U-A or A-U-G sequence in the molecule. It appears that of the two anticodons suggested for the alanine tRNA, 1-G-C and the C-G-G sequence between the two DiHU's (*1*), I-G-C is the correct one, since the region between the DiHU's in the tyrosine acceptor RNA is unlikely to be the anticodon. In serine tRNA, I-G-A has been found in a position similar to the I-G-C in the alanine RNA (*16*). G-ψ-A, then, is very probably the anticodon in the tyrosine tRNA. Since it is possible that ψ forms better hydrogen bonds than U (*17*), its presence in the anticodon should not cause any problems. It might even be beneficial (*18*).

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18. After this was written, H. G. Zachau, D. Dütting, H. Feldmann, *Angew. Chem.* **78**, 392 (1966), reported the nucleotide sequences of two very closely related serine tRNA's from brewer's yeast. Most of the discussion in our report concerning probable secondary structures also applies to the serine RNA's.
19. We thank Dr. R. W. Holley without whose encouragement and assistance this work could not have been carried out, Drs. E. B. Keller and J. Apgar for consultation, and G. Teetor and M. Simonson for technical assistance.

28 April 1966

Deep-Sea Authigenic Calcite and Dolomite

Abstract. *Crystals of calcite and, in one case, of dolomite up to 90 μm in size are dispersed in pelagic red clay at several sites on the floor of the South Pacific Ocean. They were analyzed by microscopic, x-ray diffraction, electron x-ray microprobe, and oxygen isotopic techniques. These carbonates are authigenic and were probably precipitated from hydrothermal solutions connected with deep-sea volcanic activity.*

The carbonate fraction of deep-sea pelagic sediments from the Pacific Ocean is generally composed of tests of Foraminifera and Coccolithophoridae. Such tests consist of microcrystalline aggregates of calcite; the size of the single crystals within the aggregates is usually about 1 μm or less. Larger and apparently nonbiogenous carbonate crystals are dispersed in pelagic zeolitic clays at the following locations in the South Pacific.

1) 18°35′S, 126°25′W, 4030 m below sea level (Amph 39). Bathymetric survey of an area adjacent to a sea mount was carried out by R.S. *Argo* of the Scripps Institution of Oceanography. Basaltic and hyaloclastic rocks were dredged from the sea mount, and two cores of fine, dark brown sediment were recovered from the flat area west of the mount (*1*). Both sediment samples (Amph 39 and Amph 40) consist of crystals of phillipsite, grains of hydrated basaltic glass, expandable smectitic clays, and black manganese oxide particles in addition to carbonates. The two samples differ greatly in carbonate content even though they were collected within several kilometers of each other and at similar depths. Volumetric determination gave 45.6 percent $CaCO_3$ for Amph 39 and 9.5 percent for Amph 40. Both samples contain very small amounts of microcrystalline calcitic tests of foraminifera and coccoliths. Sediment Amph 39 contains irregularly shaped, single-crystal grains of calcite up to 60 μm long (Fig. 1*a*), which are absent from sample Amph 40.

2) 18°30′S, 124°30′W, 3860 m below sea level (Amph 38). An attempt to collect a gravity core at this location resulted in recovery of only a 5-cm layer of indurated brown material in the core nose. This rock consists essentially

of grains of basaltic glass up to 200 μm in diameter (Fig. 1*b*), which in the upper few millimeters of the core are mixed with clays of the smectite group, limonitic iron hydroxides, and crystals of phillipsite and carbonates. The $CaCO_3$ content of this material is 9.8 percent; the carbonate fraction includes, in addition to coccoliths, crystals of calcite up to 90 μm in size (Fig. 1*c*), similar to those present in sample Amph 39. A few of these crystals appear to be twinned on the $(01\bar{1}2)$ plane.

3) 13°28′S, 149°30′W, 4300 m below sea level (Chall 276). Murray and Renard (*2*) described the material recovered by the *Challenger* at this station, some of which I restudied (*3*). The material includes fragments of basaltic and palagonitic rocks up to a few millimeters in size, "manganese nodules," and fine, brown, zeolitic clay containing phillipsite, volcanic glass, and smectites. Carbonate fraction of the fine sediment, mainly calcite grains similar to those of Amph 39, is 39.9 percent. A few euhedral carbonate rhombs are present, probably dolomite (Fig. 1, *d* and *e*). Some sediment grains from Chall 276 were embedded in an epoxy resin, polished, coated with a thin conductive layer of carbon, and analyzed with an Applied Research Laboratories electron microprobe. Scanning-pictures of backscattered electrons from Ca and Mg radiation from a few of these grains are shown in Fig. 2. Semiquantitative analysis of the grain in the center of the picture (*dol*) indicated a Ca:Mg ratio close to 1 and did not detect elements with an atomic number larger than 11 except for Ca and Mg. Thus, the presence of dolomite was confirmed.

Table 1. Oxygen isotope analyses of CO_2 from the carbonate fraction of deep-sea sediment samples. Carbonates treated with 100 percent H_3PO_4. The δ per mil is relative to the Chicago standard PDB-1. Temperatures calculated assuming isotopic equilibrium with present Pacific sea water.

Sample	δO^{18} (per mil)	t (°C)
Amph 39	−0.51	18.8
Amph 38 (> 25 μm)	−1.15	21.6
Amph 38 (< 25 μm)	−1.38	22.7
Chall 276	−1.22	21.9

4) 13°02′S, 104°41′W, 3780 m below sea level (Ris 56). This sediment contains 73.4 percent $CaCO_3$. The carbonate fraction consists of calcite crystals up to 80 μm long (Fig. 1*f*) and of minor amounts of foraminifera test fragments. Phillipsite and altered volcanic glass are present. Sediment containing 72.0 percent $CaCO_3$ and mineralogically similar to Ris 56 was collected at 42°00′S, 102°00′W, depth 4240 m (DWHG 48).

The calcite in the aforementioned samples is of low-magnesium type, as determined by the spacing of the (112) planes (*4*). The oxygen isotope ratios of the carbonate fraction were measured in three of the samples (Table 1). Sediment Amph 38 was split into fractions having grains of two sizes (> 25 μm and < 25 μm) prior to isotope analysis. Single-crystal grains of calcite are prevalent in the fraction with grains > 25 μm; they are mixed with calcitic coccolith aggregates in the fraction with grains < 25 μm.

There are several possible origins of these carbonate crystals in the deep sea, namely, biological processes, transport from land, or authigenous inorganic precipitation. Planktonic or deep-sea benthonic organisms do not contain large single crystals of calcite as part of their tests; a direct biological origin is thus unlikely.

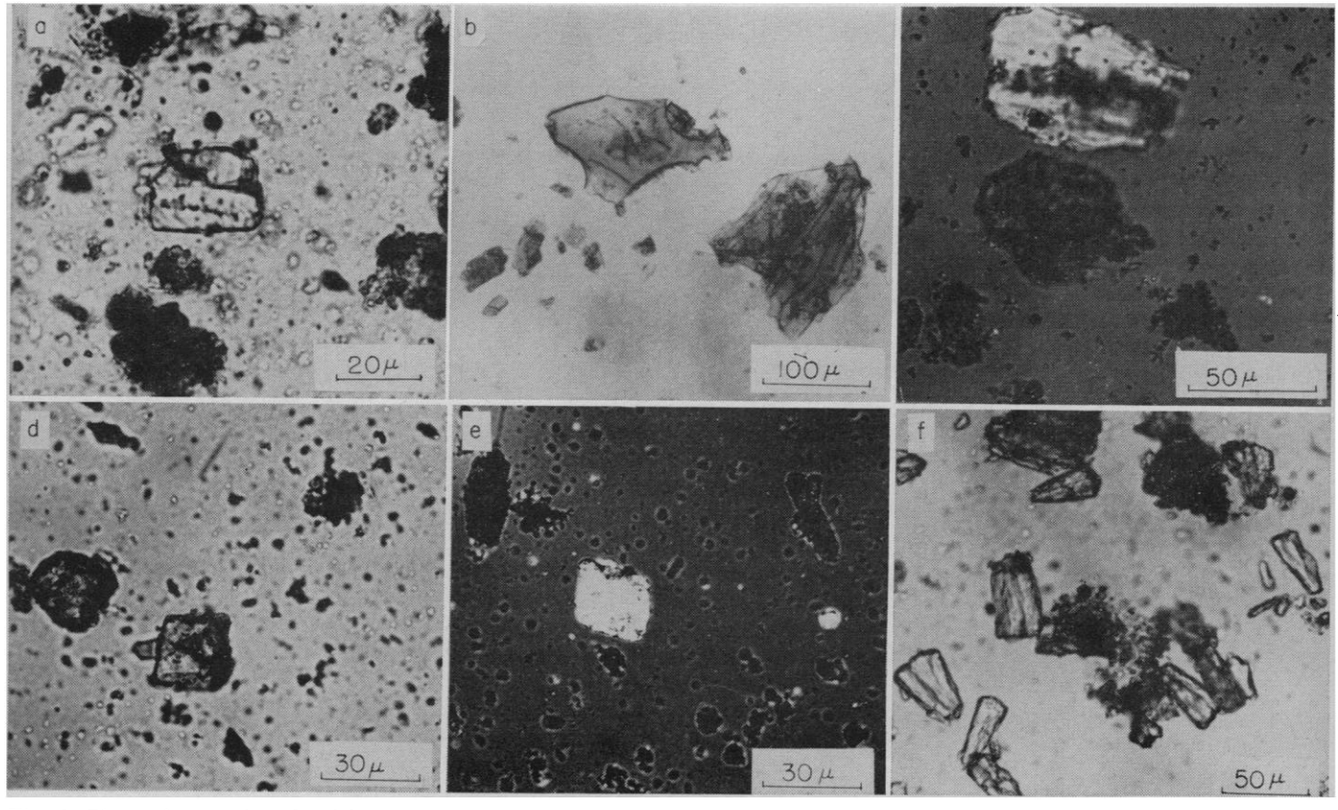

Fig. 1. Photomicrographs of calcite and dolomite crystals (transmitted light) from South Pacific deep-sea sediment: *a*, calcite crystals in Amph 39; *b*, grains of basaltic glass in Amph 38; *c*, crystal of calcite together with grains of altered glass in Amph 38 (partially crossed Nicols); *d*, rhomb of dolomite and grain of volcanic glass in sediment Chall 276; *e*, crystal of dolomite in sediment Chall 276 (partially crossed Nicols); and *f*, crystals of calcite in sediment Ris 56.

The few reported occurrences of deep-sea, nonbiogenous calcite crystals are from areas where adjacent land masses can supply calcite to the sediment (*5*). In this case no land masses are close enough to have provided a source; in fact, terrigenous minerals are quantitatively negligible in this region of the South Pacific (*6*). If these deep-sea carbonates had been transported from the continent they would be mixed with other terrigenous minerals such as quartz and micas, and their distribution on the ocean floor would tend to be homogeneous; but, in fact, it is very localized. Their large size is incompatible with having been transported from a distance.

It is generally assumed that, at the temperatures and CO_2 pressures prevailing at 4000 m below sea level, inorganic precipitation of carbonates from sea water is not possible; at or below such depths dissolution of carbonates prevails, as indicated by the rapid decrease of carbonate content with depth in Pacific bottom sediments (*7*). Precipitation of the crystals in question from normal bottom sea water seems to be ruled out by their oxygen isotopic composition. If it is assumed that the oxygen isotope ratio of the bottom water at the time of hypothetical carbonate precipitation was similar to the ratio in present-day Pacific sea water, the resulting "isotopic temperatures" of the carbonates should have been lower than those given in Table 1. Similar considerations exclude recrystallization of calcitic biogenous tests on the sea bottom as a probable mode of formation of the crystals, even though submarine recrystallization of "globigrina ooze" has been reported (*8*).

A last alternative is that the carbonate crystals under study were precipitated on the ocean floor from solutions differing in composition or temperature, or both, from normal bottom water. Each of the carbonate-containing locations coincides with an area of submarine volcanic activity. The major solid components of sample Amph 39 are alteration products of fine volcanic debris (hyaloclastites) ejected by the adjacent volcano (*2*). It is significant that the calcite crystals were found in the sediment closer to the volcano and not in the sediment several kilometers farther away. Rocks and sediment recovered at Chall 276 prove that this was a site of extensive submarine basaltic volcanism (*3, 4*). Anomalous temperature, alkalinity, and chemistry of bottom water several hundred kilometers north of station Chall 276 suggest that volatiles are being ejected on the sea floor in this area (*9*). The components of the semi-indurated rock at station Amph 38, especially the coarse grains of basaltic glass (Fig. 1), indicate that this location was also subjected to volcanic activity. The noncarbonate fraction of sediments Ris 56 and DWHG 48 consists of alteration products of volcanic glass; the samples are from the East Pacific Rise, where evidence of volcanic and hydrothermal activity is ample (*10*).

Submarine volcanic eruptions are likely to locally disturb the chemical equilibria of bottom water and also affect the carbonate system in the process. The temperature of bottom water at the eruption site will increase; consequently, some carbonate should precipitate because of the decrease of solubility of $CaCO_3$ with increasing temperature. In addition to this effect, volatiles connected with the volcanic activity may be introduced into the water. Because of the composition of magmatic volatiles, the resulting solutions should be more acidic than normal bottom sea water; therefore, they should be able to dissolve some of the biogenous carbonate ooze present on the bottom at the site of volcanic activity. Upon reestablishment of normal conditions the excess carbonate should reprecipitate in areas adjacent to the eruption. It is also possible that calcite was precipitated directly from hydrothermal volcanic solutions ejected on the sea floor. Calcite is commonly found in hydrothermal deposits on land. One of these processes, or a combination of

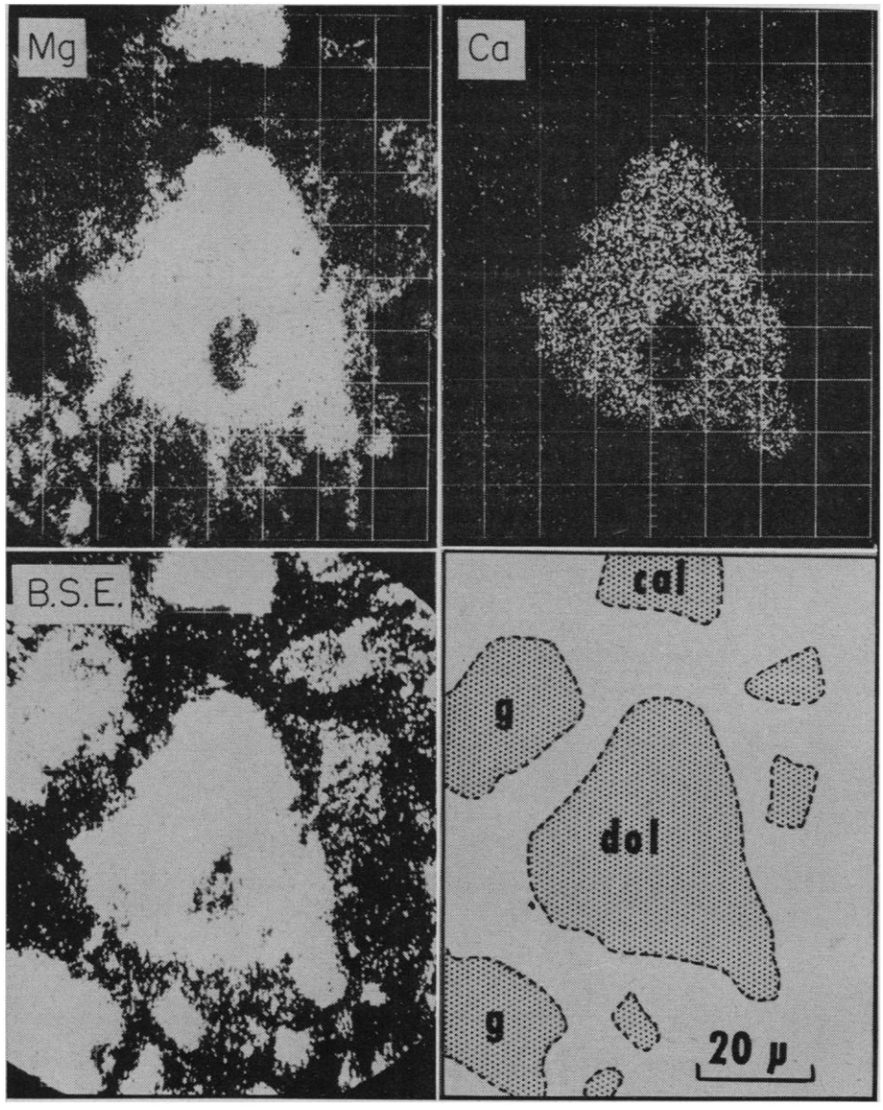

Fig. 2. Electron probe scanning pictures of back-scatter electrons (*B.S.E.*) from Ca and Mg radiation in grains from sediment Chall 276. *dol*, Dolomite; *cal*, calcite; and *g*, volcanic glass.

them, is probably responsible for formation of the authigenous calcite crystals. Such modes of formation are not inconsistent with oxygen isotope data.

Dolomite is associated with calcite in sample Chall 276. Crystals of dolomite of unspecified origin have been observed occasionally in pelagic clays of the Pacific (*11*), and a dolomite-rich layer in contact with a basalt sill has been observed in the experimental Mohole core off Guadalupe; formation of dolomite was related in that case to alteration of the basalt (*12*). Formation of dolomite in Chall 276 could be related to the same hydrothermal-type processes suggested above for the origin of associated calcite crystals. In fact, dolomite should be precipitated directly from warm solutions with a relatively high ratio of Mg to Ca (*13*).

Precipitation of carbonates as a consequence of submarine volcanic activity, based on observations of ancient, now emergent, volcanic formations, has been suggested in the past (*14*).

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3 May 1966

Biosynthesis of Gamma Globulin: Studies in a Cell-Free System

Abstract. *Incorporation of C^{14}-amino acids into high-molecular-weight material precipitable by trichloroacetic acid indicates that microsomal cell-free systems, derived from spleens of immunized rabbits, are active in protein synthesis. Protein was made soluble by ultrasonic irradiation of the cell-free incubation mixtures, and low-molecular-weight materials were removed by dialysis and gel filtration. Chromatography and radio-immunoelectrophoresis of this soluble protein fraction reveal C^{14}-labeled protein having several characteristics of gamma globulin.*

Mechanisms of the production of γ-globulin and antibody are not well defined. Immunoglobulins are synthesized primarily in the lymph nodes and spleen by lymphoid cells and plasmacytes. γ-Globulin and antibody are present in these cells (*1*), and the isolated cells produce antibody (*2*). The ribosome-rich endoplasmic reticulum of plasmacytes is thought to be the site of antibody synthesis. Sucrose density-gradient centrifugation of ribosomes isolated from spleen cells suggests that the active synthetic units are single and double ribosomes (*3*). Similar studies of lymph node homogenates have shown that either small or large aggregates may be active in protein production (*4*). Ribosomes isolated from rat spleens have been utilized in cell-free systems and they are active in polypeptide synthesis (*5*). Whereas it has been reported that γ-globulin may be identified as a product of lymph node cell-free systems by immune coprecipitation techniques (*6*), it has not, to our knowledge, been further defined (*7*). We now describe a microsomal cell-free system, derived from rabbit spleens, in which there is incorporation of C^{14}-amino acids into material having several characteristics of γ-globulin.

New Zealand white female rabbits were immunized, their spleens were removed, and the cells were separated and washed essentially as described (*3*). Microsomes and a fraction insoluble at *p*H 5 were prepared by methods used for brain cells (*8*). The fractions insoluble at *p*H 5 were also prepared from rabbit brain and liver. All fractions were frozen at −30°C and used within 24 hours of preparation. The complete incubation mixture for amino acid incorporation contained, in 0.7 ml, 25 μmole of tris, *p*H 7.4; 25 μmole of KCl; 5 μmole of $MgCl_2$; 10 μmole of glutathione; 0.5 μmole ATP (*9*), 0.125 μmole of GTP, 25.0 μmole of phosphoenol pyruvate; 0.03 mg of pyruvate kinase; 5 to 10 μc of C^{14}-leucine (233 mc/mmole); 0.1 to 0.2 mg of microsomes; and 0.2 to 0.4 mg of the fraction insoluble at *p*H 5 from rabbit spleen, liver, or brain. Larger systems, containing up to 0.5 mg of microsomes in 1.4 ml, were prepared for analytical studies. These were incubated with both C^{14}-leucine (233 mc/mmole) and C^{14}-proline (185 mc/mmole) to enhance the specific activity of the nascent protein.

Small systems, and portions of the large ones, were precipitated at 0 and 60 minutes with 5 percent TCA or a mixture of 5 percent TCA and 2.5 percent sodium tungstate. The TCA precipitates were washed (*8*) and layered on Millipore filters (0.45 μ), and radioactivity was determined in a Nuclear-Chicago gas-flow counter. The remainder of the large systems was subjected to ultrasonic irradiation for 2 to 5 minutes in a Branson Sonifier at step 6 (4.5 amp). The soluble fraction, after centrifugation at 110,000*g*, was passed through columns of G25 or G50 Sephadex; the external volume was dialyzed against ten volumes of distilled water at *p*H 5.8, with the water being changed every 12 hours for 3 days. A precipitate, which formed during dialysis, was removed by centrifugation, and the soluble material was lyophilized. In later studies, dialysis was carried out against 0.001*M* tris at *p*H 7.0 (4°C); in this case no precipitate formed, and the material was concentrated, in the dialysis tubing, against dry Sephadex G200. The lyophilized material was reconstituted in small volumes (0.1 to 0.2 ml) of appropriate buffers for chromatography and radio-immunoelectrophoresis. Samples were chromatographed on 2 g of DEAE cellulose (exchange capacity 0.68 meq/g) (*10*), and radioactivity of 0.5-ml portions of the effluent was measured in a liquid-scintillation counter with Bray's counting fluid (*11*).

Optimum incorporation of C^{14}-leucine into material precipitable by TCA is dependent upon microsomes, the fraction insoluble at *p*H 5 (enzymes), ATP, and an ATP-regenerating system. The complete system showed 7950 count/min per milligram of microsomal