

eral control experiments were run. In one experiment, 50 mg of palmitic acid was added to a sample of Baffin Bay sediment which had been extracted with organic solvents. The mixture was extracted with methanol by the use of sonification. Fatty alcohols and hydrocarbons were analyzed according to the standard procedure. No alcohols or hydrocarbons were detected. In other control experiments split samples were extracted by Soxhlet extraction and sonic energy, and the alcohol patterns were compared. In general, the total yield of alcohols was slightly higher with sonification. In qualitative composition the recovered fatty alcohols were not significantly different. The extraction procedure based on sonic energy does not create any artifacts.

Fatty alcohols in sediments probably have their origin in the marine life of the environments studied. Because Baffin Bay normally receives very little fresh water as runoff, it is often twice as saline as seawater. Such restricted runoff probably could not transport enough terrestrial organic matter to account for the uniform concentrations of alcohols observed in the bay sediments. Inasmuch as the same types and concentrations of alcohols are present in all the Recent samples, we assume Baffin Bay to be typical. We cannot specify which marine organisms are producing the normal fatty alcohols, although the bacteria may be involved. The isoprenoid alcohol, dihydrophytol, probably is derived from phytol, the side chain of the chlorophyll molecule. The reducing environment of the sediment must be sufficient to hydrogenate the double bond of phytol. Likewise, reduction of the corresponding fatty acid would yield the fatty alcohols. However, our data do not allow us to evaluate this mechanism.

Hoering (11) has isolated and identified fatty alcohols in Recent sediment while we were conducting our investigations. His results, based in part on mass spectra, are qualitatively similar to ours. The fatty alcohols promise to be a rewarding subject for organic geochemical studies. Dihydrophytol, in particular, should serve as a useful biological marker.

JUDY SEVER
P. L. PARKER

University of Texas,
Marine Science Institute,
Port Aransas 78373

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Sterile Soil from Antarctica:

Organic Analysis

Abstract. Soils from the dry-valley region of Antarctica can be sterile by the usual microbiological criteria and yet contain significant amounts of organic carbon. Examination of one such soil shows that the organic material is finely divided anthracite coal. These findings have significant implications for the biological exploration of Mars.

The ice-free valleys of Victoria Land, Antarctica, are among the world's most hostile deserts. No higher plants or animals inhabit these regions. Dense microbial populations are found where glacial melt makes liquid water available during part of the year, but the driest parts of the valleys are sterile or nearly so (1). To the extent that low temperatures and scarcity of water limit life in the dry valleys, these valleys may resemble the planet Mars; we have found these valleys useful as a model environment for the investigation of problems related to the search for life on Mars.

There is little correlation between the microbial count of dry-valley soils and their content of organic carbon as determined by the standard Allison wet-combustion method (2). Particularly anomalous are soils which show no microorganisms but which contain a significant amount of organic carbon. A possible explanation of the anomaly

is that these soils harbor an undetected population of microorganisms; we have carried out an intensive study of one of the abiotic soils in order to determine the nature and origin of the organic matter.

The soil chosen for study (No. 542) was collected during the austral summer of 1966-67 on a gentle slope facing northeast about 1.5 km west of Lake Vida, near the junction of the Victoria and McKelvey valleys. The sample was collected aseptically (3) at a depth of 15 to 25 cm and has since been stored at a temperature below -25°C . This sample was selected because it had been collected in metal or poly(chlorotrifluoroethylene) containers, rather than in standard commercial soil bags. The latter, constructed of canvas with a polyvinyl chloride liner, introduce minor amounts of solvents and plasticizers which are detectable by the sensitive method involving pyrolysis, gas chromatography, and mass spectrometry that we have developed for the organic analysis of desert soils (4, 5).

The microbial count on soil No. 542 was below the detectable limit of approximately one organism per gram in all media tested. These included the following commercially available media: trypticase soy agar, trypticase soy broth with and without added sodium chloride, lactose broth, fluid thioglycollate, Rose Bengal agar, actinomycete isolation agar, deoxycholate agar, nitrate broth, anaerobic agar with and without glucose, and Brewer anaerobic agar. The following special media were also used: simulated Taylor Valley salts (6), Thornton's algal medium (7), Burk's agar (7), Van Delden's agar (7), and yeast agar (8). Samples were incubated from 15 days to 3 months, usually at 20°C ; some samples were incubated at 2° , 25° , 37° , or 55°C . Anaerobic incubations were carried out under both N_2 and CO_2 .

Unlike some Antarctic soils which are toxic because of their high boron content (6), soil No. 542 did not inhibit growth when mixed in a 1:1 ratio with another dry-valley soil containing 1500 organisms per gram. As a further check of the sterility of soil No. 542, its capacity to generate metabolic CO_2 from labeled glucose and amino acids was determined. The results are shown in Table 1, together with comparable data for other dry-valley soils, including the entire profile (ground level to

Table 1. Microbial counts and metabolic production of radioactive CO₂ in Antarctic soils. Counts in soils Nos. 537, 540, 541, and 543 were estimated by dilution in trypticase soy broth and fluid thioglycollate; in soil No. 513 counts were estimated by plating on actinomycete isolation agar at 20°C (only bacterial colonies appeared after incubation for 30 days). Production of labeled CO₂ was measured on 300 mg of soil in 0.3 ml of water containing 144 ng of uniformly labeled glucose (5×10^4 count/min) and 20 ng of a mixture of 15 uniformly labeled amino acids (3.9×10^4 count/min). Duplicate samples were incubated for 1 hour at room temperature in sealed chambers. Reactions were stopped by addition of 0.7 ml of 5 percent trichloroacetic acid; labeled CO₂ was flushed by a stream of nitrogen into 0.2M hyamine hydroxide solution in methanol and was measured in a liquid scintillation counter at a counting efficiency of 57 percent. The drop in background counts per minute after the second set of measurements was due to a new dilution of the labeled substrate solution having been taken for the site C determinations. Net counts per minute are comparable in all experiments. Controls were sterilized by dry heat.

Soil No.	Depth (cm)	Highest dilution showing growth (g/10 ml of medium)	Estimated microbial count (per gram)	Count/min per 300 mg of soil	Net count/min
<i>Site A: McKelvey Valley north of Olympus Range, below Mt. Hercules</i>					
513 (untreated)	Top 5		7500	1577	1474
513 (sterilized)	Top 5			103	
<i>Site B: Victoria Valley, 45 m northeast of Lake Vida</i>					
537 (untreated)	Top 2.5	0.001	10^3 - 10^4	1008	899
537 (sterilized)	Top 2.5			109	
<i>Site C</i>					
540 (untreated)	Top 2.5	0.01	10^2 - 10^3	157	120
540 (sterilized)	Top 2.5			37	
541 (untreated)	2.5-15	1.0	1-10	56	-6
541 (sterilized)	2.5-15			62	
542 (untreated)	15-25	No growth	0	32	-4
542 (sterilized)	15-25			36	
543 (untreated)	25-30*	1.0	1-10	28	-33
543 (sterilized)	25-30*			61	

* Permafrost level.

permafrost) from which soil No. 542 was taken (Table 1, site C). The microbial counts from site C suggest a population in balance between bacterial fallout and mortality in an essentially abiotic environment.

Chemical analysis (Table 2) demonstrated a high nitrate content that is typical of many Antarctic soils and indicative of the absence of biological activity and leaching (9). The content of organic carbon is similar to that of some temperate desert soils containing sizable microbial populations (10). However, pyrolysis of soil No. 542 (500°C under helium) and gas chromatography and mass spectrometry of the pyrolysis products did not demonstrate volatile organic products. The same result was obtained when the pyrolysis products were passed directly into the mass spectrometer: The only volatile products produced at 500°C were carbon dioxide, nitric oxide, and sulfur dioxide. (Water was not determined, since the samples had not been rigorously dried.) These results imply an organic carbon content that is less than 5 percent of that indicated by the Allison combustion method.

Because sulfur dioxide can interfere with the Allison determination (11), soil No. 542 was analyzed for oxidiz-

able forms of sulfur. No inorganic species capable of oxidation to sulfur dioxide were detected (Table 2). The possibility that organic carbon was being oxidized to CO₂ by nitrate during pyrolysis was tested. It was found that nitrate can lead to loss of some, but not all, of the organic matter of soil. Removal of nitrate from soil No. 542 by extraction with water did not alter the pyrolysis products, except to nearly eliminate the nitric oxide peak. An independent check of the carbon analysis by a dry combustion method (12) confirmed the Allison value.

Table 2. Chemical analysis of soil No. 542. Water-soluble ions were determined in a 1:5 extract of soil to water. The pH of saturated soil paste was 7.5.

Element or ion	Percentage
C	0.29
C, organic	.11
CO ₃ ²⁻	.17
HCO ₃ ⁻ , water soluble	.01
N	.16
N, organic	.001
NO ₃ ⁻ , water soluble	.47
S	.24
SO ₄ ²⁻ , water soluble	.60
SO ₃ ²⁻	<.001
S ⁰	<.001
S ²⁻	<.001
Na ⁺ , water soluble	1.1
Cl ⁻ , water soluble	1.6
B	.0003

The elemental analysis shows an unusually high ratio of organic carbon to organic (Kjeldahl) nitrogen, an indication of a virtual absence of organic nitrogen compounds. Extractive analyses were negative for hydrocarbons and fatty acids (<0.002 percent) and for carbohydrates (<0.01 percent). Moreover, no organic infrared bands could be detected in the extracts. In view of these results, it seemed highly improbable that the organic carbon could be present in any form except as elemental carbon.

In order to verify this conclusion, carbonate and siliceous minerals were removed by alternate treatments with hydrochloric and hydrofluoric acids. The residue consisted of fine colorless granules containing black particles which disappeared on heating under oxygen. Differential thermal analysis in oxygen showed an exotherm with a maximum at 475°C, typical of finely divided carbon. Repetition of the fractionation with less vigorous agitation resulted in a mixture from which the black particles could be extracted manually. Differential thermal analysis of these particles revealed a very pronounced exotherm, with no endotherm below 450°C. No infrared or mass spectrum could be discerned. Elemental analysis indicated 66.7 percent C, 2.67 percent H, 0.27 percent N, and less than 0.002 percent S. The ratio of ¹³C to ¹²C, expressed as δ¹³C, was -24 per mil, which suggests a biological origin (13). Microscopic examination showed that some of the black particles were imbedded in a colorless mineral matrix—probably quartz—from which we conclude that the carbonaceous material is not of recent origin. Microscopic study also showed conchoidal fracturing, which is typical of coallike materials.

All of the evidence thus indicates that the organic matter in soil No. 542 is anthracite coal. Its probable source is the Mt. Bastion coal measures, about 25 km distant from the collection site. The Mt. Bastion coal is an anthracite which is described as having "no coking properties and a high proportion of ash, which could not be separated by normal methods" (14). In these respects it is indistinguishable from the carbonaceous material in soil No. 542. The chemical results thus support the conclusion that the soil is actually sterile. This sterility, which is remarkable in itself, has significant implications for planetary exploration. It sug-

gests that microbial life, if any, will not be distributed ubiquitously in harsh environments. In addition, it raises new questions regarding the vulnerability of Mars to contamination by terrestrial microorganisms (15).

N. H. HOROWITZ, A. J. BAUMAN
R. E. CAMERON, P. J. GEIGER
J. S. HUBBARD, G. P. SHULMAN
P. G. SIMMONDS, K. WESTBERG

Space Sciences Division, Jet
Propulsion Laboratory, California
Institute of Technology,
Pasadena 91103

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Precambrian Columnar Stromatolites in Australia: Morphological and Stratigraphic Analysis

Abstract. *The stratigraphic distribution in Australian Precambrian rocks of columnar stromatolites, organosedimentary structures formed by blue-green algae, has been investigated. Their morphology is being studied according to methods developed in Russia. The discovery of successive different assemblages supports not only regional but also intercontinental stratigraphic correlations which are in agreement with available isotopic datings.*

"Stromatolites stand in roughly the same relation to blue-green algae as coral reefs to corals" (1). They are sedimentary structures produced by life processes of Cyanophyta in various associations of species and genera, possibly also with other algae and bacteria. From these undisputed facts the conclusion was drawn (2) that the variable gross form of these structures was due to variations in environmental factors, although the microstructure rarely if ever shows identifiable remains of algal cells. Hence any evolutionary change which could be morphologically ascertained and lead to stratigraphic correlations based on stromatolite occurrences was considered unlikely. Correlations by means of these fossils, at ranges in time and space greater than those of specific local environments, were discouraged.

Undeterred by these views, a group of Russian workers (3, 4) carried out

systematic studies of the morphology and stratigraphic distribution of stromatolites from many regions of the U.S.S.R. From the results of their work we concluded that although the gross forms and structures of most stromatolites may be too simple to allow recognition of diagnostic characters and some variations are due to environmental factors, nevertheless, columnar forms do exhibit sufficiently clear diagnostic characters (Figs. 1 and 2). Their distribution in four successive assemblages of form genera has been convincingly described from many sequences of Precambrian rocks in the U.S.S.R. A division into five assemblages has recently been suggested (4).

We collected or obtained material from all major areas of essentially unmetamorphosed Precambrian sediments in Australia, and studied selected specimens according to Krylov's method of "graphic reconstruction" from serial

sections of columns. Illustrations of examples of form taxa and a brief discussion of most of the characters used in their distinction are given by Raaben (4). Although accepting most of these taxa, we concur with some other Russian workers in attributing more significance to gross features and less to microscopic structures which are commonly affected by diagenesis. Figure 1 illustrates terms to be used in diagnoses. Character combinations rather than single features distinguish form genera and species. Some characters vary considerably within single species, but dominance of a modal form makes it possible to recognize distinctive characters.

The results of work on two areas which is nearing completion are here reported (Fig. 3). The Skilloalee Dolomite, occurring low in the Burra Group ("Torrenian"), in the Adelaide Geosyncline, South Australia, contains a uniform assemblage of *Baicalia* spp. over a distance of 450 km. The formation typically comprises pale-colored, relatively pure dolomite and minor clastics in its lower part, and dark gray, either massive or thinly bedded argillaceous dolomites, together with sedimentary magnesite conglomerates, secondary black chert, and minor clastics in its upper part. In different localities, *Baicalia* spp. occur in each of these rock types except the clastics. Many Russian form species of *Baicalia* have been distinguished by textural features of the lamination which are at least in part diagenetic. This makes identification with previously described form species uncertain.

Conophyton cf. *garganicus* was found in a large dolomite raft in a diapir. The age of the dolomite is uncertain but it must be older than the upper part of the Burra Group, which the diapir intrudes; it probably came from the lower Burra Group or the underlying Callanna Beds. Although textures of stromatolites must be interpreted cautiously, structures (shape and thickness variations of laminae) of conophyton are distinctive and significant for the subdivision of the form genus into form species. *Conophyton* cf. *garganicus* is identified by its even dark and light laminae, the dark laminae being uniformly lenticular. *Conophyton* cf. *garganicus* occurs *in situ* in the Irregularly Formation of the Bangemall Group, in the Capricorn Ranges of the North West Division, Western Australia.

The Burra Group is everywhere over-