Bacterial Life at the Bottom of the Philippine Trench[1]

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BACTERIA IN ABUNDANCE have been found (*1*) in virtually all samples of marine bottom deposits, examined regardless of the depth of the overlying water. Until recently, however, very few samples had been taken from depths exceeding 2000 meters, and none from little more than half the depth of the most abyssal oceanic deep. Certes (*2*) reported the presence of bacteria in bottom deposits collected on the *Talisman* Expedition from depths as great as 5100 meters. Significantly, Certes (*3*) observed that the organisms from around 5000 meters were able to tolerate hydrostatic pressure approximately isobaric with this depth; namely, 500 atmospheres. On the Humboldt Plankton Expedition to the West Indies, Fischer (*4*) found bacteria in several deep-sea sediment samples, 5280 meters being the greatest depth that he explored. Marine microbiologists at the Scripps Institution recovered bacteria from mid-Pacific mud at a depth of 5300 meters (Lat. 14° 22′ N, Long. 133° 06′ W) and in a sample of bottom deposits collected off the coast of Bermuda from the floor of the Atlantic Ocean at a depth of 5800 meters.

Recently the Danish *Galathea* 'Round the World Deep Sea Expedition has afforded facilities for the collection and microbiological analysis of sediment samples from the deepest known parts of the oceans. Many samples were taken from depths exceeding 6000 meters, including several from the Philippine Trench at depths greater than 10,000 meters. Confirming and further delineating the observations of the U.S.S. *Cape Johnson* (*5*), this trench was shown by echo depth recorder to extend for about 100 miles from 9° 04′ N × 127° 05′ E to 10° 41′ N × 127° 37′ E on a line about 20° west of north. For most of its length its walls rise precipitously from the bottom, which is only from one-half mile to three miles wide. Only by the best of navigation was it possible to delineate the trench and to stay on station from 8 to 18 hours, the time required to collect a sample from the bottom. Often this was achieved in the face of adverse meteorological conditions characteristic of the monsoon and typhoon season in Philippine waters during July and August 1951.

Samples of bottom deposits were collected by means of a plastic-lined Kullenberg corer and a Petersen grab. Immediately after they were brought to the surface, radially central samples were removed with sterile instruments to secure uncontaminated material for microbiological analysis. The direct microscopic method and several standard cultural procedures were employed to demonstrate the presence of bacteria. Duplicate cultures were incubated in the ship's refrigerator at 2.5° C, which approximates the sea floor temperature, and at air temperature, which varied little from 30° C. Part of the cultures were incubated at atmospheric pressure and part in steel cylinders (*6*) at approximately the pressure of the environment from which the inocula were collected. Representative findings for five samples from the Philippine Trench are summarized in Table 1. For comparison the results from two samples taken from lesser depths on either side of the trench are presented. Sea water enriched with peptone, beef extract, and yeast extract was employed as the culture medium.

Table 1 shows the number of bacteria demonstrated in the topmost layer of bottom sediments by the minimum dilution method. The abundance of bacteria was found to decrease sharply with depth below the mud-water interface. Only 10 to 100 viable cells could be detected per gram of mud from depths 75 to 100 cm below the surface of the sea floor. All cultural counts must be regarded as minimal, however, because no one medium or set of cultural conditions could conceivably provide for the reproduction of all bacteria.

The presence of large numbers of bacteria in bottom deposits was confirmed by the direct microscopic observation of freshly collected samples. Direct microscopic counts, however, had little quantitative significance, owing to the difficulty of distinguishing particles of sediment from bacteria, and other practical difficulties caused by vibration and other movements of the ship at sea.

The largest cultural counts were obtained in nutrient medium incubated at around 2.5° C. It is also

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TABLE 1

NUMBER OF BACTERIA PER GRAM WET WEIGHT OF BOTTOM DEPOSITS DEMONSTRATED BY THE MINIMUM DILUTION METHOD

Station No.	Location of station		Water depth (meters)	Incubated at *ca* 30° C		Incubated at *ca* 2.5° C	
	Latitude	Longitude		1 atm	1000 atm	1 atm	1000 atm
413	10° 20′ N	126° 36′ E	10,387	10^2	10^3	10^4	10^5
418	10° 13′ N	126° 43′ E	10,462	10^2	10^3	10^3	10^6
419	10° 19′ N	126° 39′ E	10,417	10^3	10^3	10^3	10^5
420	10° 24′ N	126° 40′ E	10,418	10^3	10^5	10^4	10^5
421	10° 29′ N	126° 05′ E	1,023	10^4	0	10^5	0
422	10° 49′ N	126° 01′ E	2,010	10^4	0	10^6	0
424	10° 28′ N	126° 39′ E	10,395	10^3	10^4	10^4	10^6

noteworthy that from ten to a hundred times more bacteria from the bottom of the Philippine Trench developed at 1000 atmospheres than at atmospheric pressure. Such observations establish the occurrence of barophilic bacteria at the greatest known oceanic depths, but much more work will be required to explain the temperature-pressure relationships of the organisms.

The time required (4 to 10 hours) to haul the samples from the deep-sea floor to the surface may account for finding in the mud no abnormally elongated bacterial cells such as those observed by ZoBell and Oppenheimer (*6*) in cultures incubated at 600 atmosphere. Predominant were rod-shaped bacteria ranging in length from 2 to 4 μ. Many were capsulated. Endospores were found in most mud samples. Pairs of rods were common, and some occurred in short chains. Species of *Pseudomonas, Flavobacterium, Vibrio, Spirillum, Bacterium, Micrococcus, Bacillus,* and *Clostridium* were identified, and probably other genera were present. The identification of species poses special problems because of the unique pressure and temperature requirements of the deep-sea bacteria.

It may be more remarkable that many bacteria from abyssal depths survived at 1 atmosphere and 30° C than that most deep-sea bacteria seemed to prefer the low temperature and high pressure characteristic of what appears to be their natural habitat. Perfected sampling techniques will be required to determine to what extent deep-sea bacteria may be injured by the sudden changes in pressure and temperature incidental to their transfer from the sea floor to the surface. Finding some survivors fails to prove that many others may not have succumbed to the sudden change of climate. None of the animals recovered from the bottom of the Philippine Trench reached the surface alive. As reported by Bruun (*7*), the bottom fauna included actinians, amphipods, bivalves, echiurid worms, holothurians, and tanaids. The author concurs with Bruun in believing that deep-sea bacteria as well as animals are affected more by the change in temperature than by the change in pressure, although the two effects are closely related. As indicated by the observations of ZoBell and Johnson (*8*), who applied the term "barophilic" to bacteria having a preference for high hydrostatic pressure, the temperature tolerance of bacteria is affected by the pressure.

Barophilic bacteria in abundance have also been demonstrated in several other abysses at depths ranging from 7200 to 10,080 meters, including the Soenda Deep in the Indian Ocean, the Weber Deep in the Banda Sea (*9*), and the Kermadec Deep in the south Pacific Ocean (*10*). Besides functioning as geochemical agents the deep-sea bacteria are believed to serve as an important source of food for bottom-dwelling animals, whose only other source of nutrient is the sparse supply of organic substances reaching them from the photosynthetic zone. The bacteria, which are known to nourish many animal species, can obtain their energy for growth from waste or dissolved organic matter in the circulating sea water or, in the case of certain autotrophic bacteria, from the oxidation of ammonium, methane, hydrogen, and possibly other inorganic substances.

Besides establishing the existence of bacterial life on the sea floor at depths exceeding 10,000 meters, the greatest depth being 10,462 meters, the project has provided barophilic cultures for further investigations of pressure as a physiological factor.

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