

Marine bacteria

Recollections and problems

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EARLY IN the spring of 1931 I received a telephone call from the venerable biologist, Professor EDWIN CONKLIN of Princeton University, to the effect that Professor HENRY BIGELOW of Harvard was visiting him that day. They were discussing organizational plans for the newly established Oceanographic Institution at Woods Hole, Mass. The question had come up as to whether it would be desirable to establish a project in marine bacteriology at the Institution. Would I care to come to Princeton and present my ideas concerning the potentialities in this field of research?

I could lay claim to only very limited knowledge of marine bacteria. What little knowledge I had was based on a certain familiarity with a few problems in which marine bacteria were involved. One of these was concerned with precipitation of lime in sea-water. Just prior to World War I, a British bacteriologist, Dr. G. H. DREW, made a study of the precipitation of lime in tropical and subtropical water, notably in the region of the Bahamas. DREW reported this process to be a result of bacterial action. Certain groups of bacteria concerned with the reduction of nitrate to atmospheric nitrogen, or the so-called "denitrifying" types, were said to be primarily involved in the process. The untimely death of Dr. DREW in 1913 brought an end to these investigations.

Seven or eight years later, Professor CHARLES LIPMAN of the University of California became interested in this problem. Together with an assistant, he spent two or three summers in the laboratory of the Dry Tortugas, in an attempt to confirm DREWS' results. Prof. LIPMAN visited me on several occasions and we had an opportunity to discuss this problem in detail. It appeared to me that the final answer had not been reached, certainly not as regards the role of bacteria in the process.

In 1929, a group of geologists from Princeton, headed by Professor RICHARD M. FIELD, organized an expedition to the Bahamas, particularly Andros Island, for the purpose of studying in detail this precipitation problem. Since "drewite", the name given to the lime formation, was believed to be of bacteriological origin, it was felt that it would be desirable to have a bacteriologist participate. I was invited, but declined, because neither my available time nor my scientific interests, I felt, would permit me to do justice to the study. I was at that time on the point of developing a comprehensive study of "organic matter decomposition by micro-organisms and of humus formation", and could not, therefore, afford to undertake a new problem that would require at least several months of my time. The Princeton group decided to invite a qualified bacteriologist interested in this problem. On my part, I promised to make facilities available in my laboratory for this study.

In 1930, Dr. W. BAVENDAMM, a German bacteriologist, came to this country to spend six months working on this problem. He was immediately placed in our

laboratory, where he was supplied with the necessary glassware and chemicals. We also helped to equip him with all the other materials required for the bacteriological phase of the expedition and discussed various aspects of the problem and the methods of approach. It was decided to limit the field expedition to the collection of samples and to carry out the studies themselves in our laboratory. Upon his return from the expedition, Dr. BAVENDAMM began to examine bacteriologically the samples of seawater and sea bottom material.

Upon studying the data, I was struck by the frequent occurrence of a certain kind of bacterium which brought about rapid liquefaction of the agar in the medium. Each colony of this bacterium was surrounded by a clear, saucer-like circle of the liquefied agar. Because of my interest in organic matter disintegration, including such compounds as hemicelluloses and polyuronides, of which agar was a type, I suggested to BAVENDAMM that we both undertake a detailed study of this bacterium and its mode of action upon the agar. He agreed, and the remaining months of his stay in my laboratory were devoted to this investigation. Our results were incorporated in a joint paper, which was submitted to the *Journal of Bacteriology*, and BAVENDAMM himself published a paper on the question of calcium precipitation.

It was just about this time that the Oceanographic Institution was being organized. The director, Dr. HENRY BIGELOW, was approached by Dr. FIELD, who was requesting support for another expedition to the Bahamas. When asked about the accomplishments of the first expedition, Dr. FIELD cited the above two papers. When Dr. CONKLIN's opinion was asked about this matter, he suggested that I be consulted. Hence the telephone call to which I have referred.

There was another reason for consulting me on this matter. Since any comprehensive survey of the field of marine bacteriology would involve an understanding not only of bacteriological processes but of complex bacterial or even microbial populations, a prior knowledge of other complex populations would be helpful. Inasmuch as my own field of study, that of soil microbiology, involved populations and relationships, there was a parallelism between the two. This could be contrasted to medical and industrial microbiology, where single cultures of organisms are concerned. Further, the cycles of life in both the soil and the sea are similar, both as regards the activities of various specific groups of micro-organisms, such as nitrifying, denitrifying, and cellulose-decomposing types, and the bacterial population of the natural substrate as a whole.

We spent several hours that beautiful spring afternoon in Princeton, in 1931, discussing various problems bearing upon marine bacteriology. All three of us reached the conclusion that neither oceanography nor marine bacteriology would gain much from another expedition to the Bahamas. However, we all felt strongly that a definite place on the research program of the Oceanographic Institution should be given to bacteriological investigators. Dr. BIGELOW then suggested that I present a tentative plan for such a program. Within a few weeks I submitted a plan based on the idea that one or two investigators interested in complex microbiological activities be invited to spend two or more months every summer at the Oceanographic Institution. A laboratory was to be set aside for this purpose, and several assistants assigned to the project. Further, the senior investigator would continue during the rest of the year to work on one of the problems in his own laboratory, assisted by one or more graduate students provided by the Oceanographic Institution.

My plan was accepted by the Trustees of the Institution, and I was invited to spend a month that summer at Woods Hole to study the situation a little more closely. Subsequently, I was appointed investigator to organize the work in marine bacteriology.

The next 10 years found me busy every summer for about two months at the Oceanographic Institution, searching for bacteria in the sea. There were always several collaborators, assistants, and graduate students to help in the research program. It is of particular interest to mention among the associates Dr. CORNELIA CAREY, Professor of Bacteriology at Barnard, Dr. MARGARET HOTCHKISS of New York Medical College, and, later, Dr. AUSTIN PHELPS of Yale University. Among the assistants who should be mentioned are, first, HERBERT W. REUSZER, CHARLES RENN, J. STOKES, and D. Q. ANDERSON, and, later, CHARLES WEISS, DONALD REYNOLDS, and DON JOHNSTONE. There were also various visiting investigators concerned with microbiological problems, principally Prof. H. GRAN of Oslo, Norway, who has left his mark in bacteriology with his classical study of the agar-liquefying bacteria; Dr. FREDERICK K. SPARROW, who made a study of the fungi in the sea, especially those pathogenic to marine algae; and Prof. U. VARTIOVAARA of Finland. A number of summer laboratory assistants, especially ERIC WARBASSE, contributed much in collecting equipment, for both inside and outside (or boat) work, and helping to organize the laboratory at the Institution.

By the time I started the first investigation in 1931, the laboratories were fully equipped and ready to operate, thanks to the efforts of REUSZER, who did a great deal of the preparatory work, especially prior to my arrival at Woods Hole. He also continued some of the experimental studies in New Brunswick during the rest of the year. It was not until 1932, however, that our program of marine research was fully developed. During the preparatory period I had full opportunity to familiarize myself with the literature on the subject. We decided to approach the study of marine bacteria along four distinct lines:

1. The nature of the bacterial population as a whole, both in sea-water and in the sea bottom. Special attention was given to the influence of distance from land, depth of water, and nature of bottom material. A comparative study was also undertaken of the methods to be used, including suitable media, and of the changes in the population after the samples were taken, especially upon standing in the laboratory. Although most of these studies were quantitative in nature, others were also qualitative, since they involved a study of the specific nature of marine bacteria, such as aerobic *vs.* anaerobic, and spore-forming *vs.* non-spore-forming types.

2. The specific nature of some of the marine bacteria. Particular attention was paid to those responsible for the formation of nitrite and nitrate by oxidation of ammonia, the reduction of nitrate, the fixation of nitrogen, the decomposition of cellulose, and the nature and decomposition of marine algal constituents.

3. Transformation of organic matter in the water and in the bottom material. This included a variety of reactions, such as oxygen consumption, carbon dioxide liberation, and nitrogen transformation. The water from warm regions showed, for example, a higher bacterial population with much less oxygen consumption than did the water from cold regions, when samples of water were kept under identical conditions for equal periods of time. This pointed to a greater concentration of available organic matter in the colder waters.

4. Various other problems came up for consideration during these years. The most important were the following:

(a) A study of the agents responsible for the disappearance of eel-grass along the Atlantic shore. RENN, who was appointed the senior assistant in marine bacteriology after the resignation of REUSZER, undertook this task and made a notable scientific contribution to it.

(b) A study of the presence or development in the sea of bacteria antagonistic or destructive to other bacteria. These particular investigations came toward the end of my stay in Woods Hole, in 1941 and 1942, and were influenced largely by my major interests at Rutgers on the antagonistic interrelations among micro-organisms and the production of antibiotic substances.

(c) The role of bacteria in the fouling of ship bottoms. This problem also came toward the end of my work at Woods Hole and on the eve of World War II. Since the chemists and biologists of the Institution soon took over this problem, it will no doubt be reported in this volume in further detail.

In addition to my work in the laboratory at the Oceanographic Institute, limited periods were spent elsewhere in connection with some of the marine microbiological problems. It is sufficient to mention two brief stays at the Bermuda Biological Station, and various trips on the ocean-going laboratory vessels, such as the *Atlantis*. The first were successful, since I had a first-hand opportunity to compare the bacterial population and the bacteriological changes in the waters in a warm region with those of cold regions. The ocean trips, however, were a complete failure, so far as I was concerned. Under the threefold movements of the ship, I immediately became seasick and had to leave the work largely to one of the assistants who always accompanied me. After two valiant efforts to carry out studies on the moving boat, I made no further attempts. Dr. BIGELOW's comment, "Food was wasted on him", was fully justified, and I gave up further sea voyages for bacteriological exploration. Fortunately, some of the assistants, especially RENN, REYNOLDS, and JOHNSTONE, were excellent sailors, and could take good care of the problems under consideration.

Thus came to an end a decade of exploration of the sea for marine bacteria. The following list of papers published from the Oceanographic Institution bears evidence of the scope of the subject, the variety of problems involved, and some of the results attained. It affords proof of the vision and far-sightedness of the founder of the Oceanographic Institution and its first Director, Dr. HENRY BIGELOW, to whom this note is gratefully dedicated.

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