Professor Bain will spend six weeks at Marble, Colorado, investigating the fabric of the Treasury Mountain granite and its roof rocks. This study is expected to demonstrate the effect of recrystallization upon petrofabric of quartz, calcite and mica in a variety of rocks, the relation of fabric and grain stability to distance from the intrusive, the areal distribution of strain around a plutonic intrusive and the relationship between contact metasomatism and strains. Data on strains in the granite and the adjacent roof may be expected to yield data bearing upon viscosity of magmas. $300.

Charles C. Mook, American Museum of Natural History, New York, N. Y. Dr. Mook will continue the work on his monograph on the Fossil Crocodilia of the World. This work has been under way for a number of years and will be brought to completion during the year. $1,500.

David Griggs, Jefferson Physical Laboratory, Harvard University. Dr. Griggs's investigation of the deformation of rocks by low stresses acting over long periods of time has been going on for two years. The laws of dry deformation have already been outlined. New experiments on specimens deformed while immersed in solutions show marked differences in behavior. New "creep testers" provided for under this grant will permit further investigation of recrystallization flow. $1,200.

Horace G. Richards, New Jersey State Museum, Trenton. Dr. Richards will join forces with Professor H. H. Hess, of Princeton University, on an expedition to the Island of Margarita off the eastern Venezuelan coast, there to collect and study Pleistocene and Recent mollusks. His work will be of immediate value to Professor Hess in his studies of the very large negative gravity anomalies of the vicinity and will contribute further knowledge of the paleogeography of the Caribbean region. $300.

SPECIAL ARTICLES

ON THE PRESENCE OF AZOTOBACTER AGILIS IN AMERICA

BEIJERINCK[1] created the genus *Azotobacter* for the non-symbiotic nitrogen-fixing aerobic bacteria discovered by him in 1901. Two species were suggested; namely, *Azotobacter chroococcum,* a soil organism, and *Azotobacter agilis,* isolated from water. Lipman,[2,3] in America, described two additional species: *Azotobacter Vinelandii* and *Azotobacter Beijerinckii.* The former is related to *Az. agilis* by the formation of a greenish pigment, while the latter is similar in certain characters to *Az. chroococcum.* All four species are recognized at the present time. In addition, other species reported in the literature are *Azotobacter Woodstownii* by Lipman[3] and *Azotobacter vitreum* by Löhnis and Westermann.[4] These, however, generally are not recognized as well-defined species.

Of all the above *Az. chroococcum* is considered the most typical species of the group, and apparently it has world-wide distribution in soil. *Az. Vinelandii* and *Az. Beijerinckii* have been reported also as present in soil, although not so frequently as the former species. It was first believed that the presence of *Az. agilis* was restricted to the canal water of Delft, Holland, from which the original isolation was made by Beijerinck. The Beijerinck strain being lost, Kluyver and van Reenen[5] isolated an organism identical with *Az. agilis* excepting that there was a lack of pigment formation. Later, Kluyver and van den Bout[6] isolated a pigment-forming strain, and considering it typical of *Az. agilis,* they named the previous isolation *Azotobacter agilis* var. *atypica.*

Recently, the extensive investigation of Winogradsky[7] reported the isolation of this species from surface waters in France. Many characteristics useful in the identification of the species were included in this publication. This isolation, together with the present report, indicates that *Az. agilis* may be distributed more widely than was supposed previously.

In the present investigation, begun in early July of 1938, the technique of Winogradsky was followed using several samples of surface water in 100 cc amounts of different origins at Madison, Wisconsin, and, later, from San Francisco, California. In addition to the original medium recommended by Winogradsky, in which ethyl alcohol is the source of carbon, a slight modification of this was included in which Fe citrate was substituted for Fe chloride.

As shown in Table 1 five samples of water and one sample of sewage effluent were positive out of twenty-two samples examined. The advantage of the modified medium is shown in samples 1, 3 and 5, in which growth occurred only in the modified medium and also in samples 2 and 4, in which cases growth occurred first in the modified medium. Positive samples were characterized by a peculiar purple or violet coloration, especially in the modified medium.

The strains isolated although similar, may be divided into two groups. The strains isolated from lake water are characterized by the lack of pigment and scanty growth in solid mediums, and the production of a

[1] M. W. Beijerinck, *Centralbl. f. Bakt.,* II Abt., 7: 561–582, 1901.

[2] J. G. Lipman, N. J. Agr. Exp. Sta. Ann. Report, 24: 217–285, 1903.

[3] *Ibid.,* 25: 237–289, 1904.

[4] F. Löhnis and T. Westermann, *Centralbl. f. Bakt.,* II Abt., 22: 234–254, 1909.

[5] A. J. Kluyver and W. J. van Reenen, *Archiv f. Mikrobiol.,* 4: 280–300, 1933.

[6] A. J. Kluyver and B. T. van den Bout, *Archiv f. Mikrobiol.,* 7: 261, 1936.

[7] S. Winogradsky, *Ann. Inst. Pasteur,* 60: 351–400, 1938.

TABLE 1
COMPARATIVE ENRICHMENTS FOR *Azotobacter agilis* WITH WINOGRADSKY'S AND MODIFIED MEDIUMS

No.	Sources of the water	Original W. medium	Modified medium	Observations
1	Lake Mendota (Madison, Wisconsin) sample No. 1	–	+	.....
2	Lake Mendota (Madison, Wisconsin) sample No. 2	+	+	First growth in modif. medium
3	Lake Mendota (Madison, Wisconsin) sample No. 3	–	+	.....
4	Sewage effluent (Madison, Wisconsin)	+	+	First growth in modif. medium
5	Lower Crystal Spring Reservoir (San Francisco, Cal.)	–	+	.....
6	San Andres Reservoir (San Francisco, Cal.)	+	+	.....

Total number of samples examined for the presence of *Az. agilis:* 22.

faint gold or purple pigment in liquid mediums depending on the nature of the carbon source. In contrast to these, the strains isolated from sewage produced a definitely greenish pigment and grew better in agar mediums. Pigment production in liquid mediums was stronger than in the other group of cultures. The strains of both groups are very motile, and in general the morphological and physiological characters agree with those of the descriptions by the previous investigators. The size of the cells is 2.4–2.8 × 2.5–4.5 μ (taken from pictures originally magnified 200 ×). The cultures used for measurement were grown on Winogradsky's medium with 1 per cent. agar and 0.5 per cent. ethyl alcohol, and a small amount of calcium carbonate. No cultures grew with the use of mannite as a source of carbon, either in liquid or in solid mediums.

For comparison a culture of *Az. Vinelandii* from the Department of Agricultural Bacteriology of the University of Wisconsin was included in this study. Morphologically it differs from *Az. agilis* strains by having elongated cells (1.4–1.6 × 2.5–3.5 μ), usually in pairs, and it is less actively motile. In contrast to the *Az. agilis* the strain of *Az. Vinelandii* grew readily on both solid and liquid mediums with mannite as a source of carbon, producing a greenish fluorescent pigment.

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LIVER EXTRACT AS A SUBSTITUTE FOR SERUM IN THE CULTURE MEDIUM FOR ENDAMOEBA HISTOLYTICA[1]

THE first practical method for the cultivation of *Endamoeba histolytica* was published by Boeck and Drbohlav[2] in 1925. The medium consisted of a solid egg slant overlaid with a liquid composed of eight parts sterile Locke's solution and one part of sterile human blood serum. The following year Dobell and Laidlaw[3] used horse serum instead of human serum in the liquid portion of the medium and demonstrated that the addition of sterile rice starch produced more abundant growth of the amoebae and prolonged the life of the cultures, thus requiring less frequent transplants. This medium is used extensively at the present time in the cultivation of *E. histolytica.* Although many suggestions for the improvement of this medium have been made, they have consisted of changes only in the solid portion of the medium. The liquid portion of the medium has consisted in all cases of dilutions of human or animal serum or egg albumen, the most widely used being horse serum-Ringer (1–6). Numerous substitutes have been tested, notably by Cleveland and Collier,[4] but without success.

During the past six months we have been using a 0.5 per cent. solution of liver extract in an 0.85 per cent. solution of sodium chloride as a substitute for horse serum-Ringer. The results obtained have been fully as good as with the serum medium, and the advantages of liver extract in both experimental and diagnostic work are many.

The liver extract which we have used most extensively is Lilly's liver extract No. 343, which is employed in the treatment of pernicious anemia.[5] The powdered commercial product is dissolved in normal saline and sterilized in an autoclave at 15 pounds pressure for 30 minutes. The solution need not be filtered, as there is very little sediment. The solution of liver extract is then added to the sterile solid medium together with a small amount of sterile rice flour. The medium is then tested for sterility by incubating for 24 hours, and is stored in the refrigerator until used.

[1] Assisted by a grant from the Division of Medical Sciences of The Rockefeller Foundation.

[2] W. C. Boeck and J. Drbohlav, *Amer. Jour. Hyg.*, 5: 371–407, 1925.

[3] C. Dobell and P. P. Laidlaw, *Parasitology,* 18: 283–318, 1926.

[4] L. R. Cleveland and J. Collier, *Amer. Jour. Hyg.*, 12: 606–613, 1930.

[5] Kindly furnished for experimental purposes by Eli Lilly and Company.