PRECIPITATION OF FERRIC HYDRATE BY **IRON BACTERIA**¹

INFORMATION concerning the physiology of iron bacteria is still very incomplete, and as a consequence there is confusion as to whether or not any of the bacteria which are encountered in iron-bearing waters are concerned directly or indirectly in the precipitation of iron from the waters. Even if it is assumed that some of the bacteria are responsible for the precipitation of ferric hydrate, it is difficult to establish with certainty the reactions whereby the precipitation is effected.

Few of the iron bacteria have as yet been cultivated in artificial media,^{2, 3, 4, 5, 6, 7} and in only a few instances is there more than meager evidence that the bacteria oxidize inorganic ferrous iron to ferric iron and utilize the energy thus liberated for growth. On numerous occasions slimy filamentous material which appears in water basins has been referred to as "iron bacteria," even though there may have been relatively little ferric hydrate associated with the cell material.

There is reason to believe that some of the iron bacteria are strict autotrophs, particularly the species of Gallionella. Some of the other iron bacteria such as certain species of Leptothrix and Crenothrix may be facultative autotrophs.⁵ Still other bacteria, which should not be called "iron bacteria," precipitate iron as a result of the decomposition of organic iron compounds. With the bacteria of the last group the iron plays no role of particular importance in the development of the bacteria, since the energy that the bacteria utilize is obtained by the oxidation of the organic portion of the compounds.

According to Winogradsky's concept of iron bacteria,^{8, 9} only bacteria that are able to grow from the energy liberated by the oxidation of inorganic compounds of ferrous iron to ferric hydrate should be called "iron bacteria." There is still considerable uncertainty as to how many genera and species would qualify as iron bacteria according to this definition. Although many other organisms are concerned with the precipitation of ferric hydrate, all the iron bacteria are characterized by the accumulation of an

¹ Journal Series Paper of the New Jersey Agricultural Experiment Station, Rutgers University, Department of Microbiology.

² H. Molisch, "Die Eisenbakterien." Jena: Gustav Fischer. 83 pp. 1910.
³ R. Lieske, Jahr. f. wiss. Botanik, 49: 91-127, 1911.
⁴ R. Lieske, Cent. Bakt. etc. II, 49: 413-425, 1919.
⁵ N. Cholodny, "Die Eisenbakterien." Jena: Gustav

Fischer. 162 pp. 1926. ⁶ M. S. Cataldi, *Folia Biol.* (Buenos Aires), October-

December, 1937-38, Nos. 79-82. 7 M. S. Cataldi, "Estudio fisiologico y sistematico de

algunas Chlamydobacteriales." Thesis, University of Buenos Aires, Argentina. 96 pp. 1939. ⁸ S.Winogradsky, Bot. Zeit., 46: 262-270, 1888.

9 S. Winogradsky, Cent. Bakt., etc., II, 57: 1-21, 1922.

abundance of ferric hydrate. The reaction by which they are presumed to grow yields very little energy, and, since the product of their oxidation is insoluble, voluminous accumulations of ferric hydrate characterize their development. Winogradsky stated that there might be as much as 100 times as great an amount of ferric hydrate as cell material of the bacteria.⁹ The following calculations suggest that this is a conservative estimate.

The reaction which characterizes these bacteria is the following:

 $4 \text{FeCO}_3 + \text{O}_2 + 6 \text{H}_2\text{O} = 4 \text{Fe}(\text{OH})_3 + 4 \text{CO}_2 + 40 \text{ Cal.}$

According to this reaction 10 calories are released for each gram atom of ferric carbonate oxidized or for 55.8 g of iron in the form of ferrous carbonate.

It has been found that the efficiency of the conversion of bicarbonate carbon to cell material by various autotrophic bacteria varies between 5 and 10 per cent.¹⁰ That is, only 5 to 10 per cent. as much energy is represented in the organic compounds of the bacterial cells as is released by these organisms during their growth on the energy obtained by the oxidation of their specific inorganic sources of energy. The efficiency of Thiobacillus thiooxidans, which has been studied more thoroughly than that of any of the other autotrophic bacteria,^{11, 12} has been calculated to be about 8 per cent.^{10, 11}

It requires 690 calories to produce one gram molecule of glucose from carbon dioxide and water or 115 calories to transform one gram atom or 12 grams of carbon to cell material according to the following reaction:

 $6CO_2 + 6H_2O = C_6H_{12}O_6 + 6O_2 + 690$ Cal.

It may be assumed, without greatly affecting the accuracy of the calculations, that the organic material of the cells is glucose. Since the cell material is somewhat more highly reduced than glucose, slightly more energy would be required than that indicated by the reaction whereby glucose is synthesized from carbon dioxide and water.

As stated above, 10 calories are liberated by oxidation of 55.8 g of iron in the form of ferrous carbonate. If the process of carbon assimilation is only 8 per cent. efficient, only 0.8 calories would be represented by the organic material in the cells of the bacteria produced by this oxidation. The amount of carbon assimilated by this amount of energy would be 0.8

 $\frac{0.0}{115} \times 12 = 0.0835$ g.

Assuming organic material similar to that of carbohydrate, the amount of organic material represented

10 L. G. Baas-Becking and G. S. Parks, Physiol. Rev., 7: 85-106, 1927.

¹¹ R. L. Starkey, Jour. Bact., 10: 135-163; 165-195, 1925.

¹² R. L. Starkey, Jour. Gen. Physiol., 18: 325-349, 1935.

by this amount of carbon would be $\frac{5}{2} \times 0.0835 = 0.2088$

g. Thus, from the oxidation of 55.8 g of iron as ferrous carbonate, there would be formed 106.8 g of ferric hydrate but only 0.209 g of organic cell material. The ratio of the weight of ferric hydrate to the weight of the cell material would thus be about 500 to 1. It is apparent that if these bacteria have efficiency similar to that of other autotrophic bacteria the organic cell material will be only a fraction of the amount of ferric hydrate produced by the oxidation process. Even assuming that the organic cell material contains a greater percentage of water than that of the ferric hydrate and allowing for the higher specific gravity of the ferric hydrate, the volume of the ferric hydrate would be many times as great as that of the organic portion of the bacterial cells.

It can be concluded that, under conditions where iron bacteria grow at the expense of the oxidation of ferrous iron, there will be large quantities of ferric hydrate. It seems unreasonable, therefore, to conclude that an organism is an iron bacterium or that it is developing as an iron bacterium unless there are far greater quantities of ferric hydrate than cell substance in the accumulated materials resulting from bacterial growth. There is reason to conclude that, where most of the products of bacterial development consist of organic filamentous material and slimes, these bacteria have grown upon organic materials instead of ferrous iron.

There are so many reactions whereby iron may be precipitated from iron-bearing waters without the participation of iron bacteria^{13, 14} that it is not safe to conclude upon the basis of iron precipitation alone that the product was formed by iron bacteria. Although not all cases of iron precipitation from water are due to iron bacteria, an abundant precipitation of ferric hydrate can be expected whenever the iron bacteria are growing by the oxidation of inorganic ferrous compounds.

There is great need for additional information on the physiology of the iron bacteria, and until more specific information is obtained it seems certain that there will be confusion regarding the identity of iron bacteria, the reactions with which they are concerned and their importance in the precipitation of ferric hydrate in nature.

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SHORT AND LONG FOOD CHAINS AMONG **VEGETABLE CROPS**

"LENGTH of food chain" refers to the number of 13 H. O. Halvorson, Soil Sci., 32: 141-165, 1931.

14 R. L. Starkey and H. O. Halvorson, Soil Sci., 24: 381-402, 1927.

animal organisms that have consumed the original food produced by plants before it is used in human metabolism. The consumption of plants would be considered a "short chain," that of meat and fish a "long chain," since animals must live on organic material produced originally by plants. Our vegetable plants seem to show an analogous relation when one compares the efficiency of those that furnish a vegetative part for food and those that furnish a fruit part. Thus, our fruit crops are those in which fertilization usually occurs before the eatable portion is produced. Vegetative crops are those where the root, stem, leaf and immature flower parts (cauliflower, broccoli and globe artichokes) are consumed.

Cabbage, carrots, potatoes, spinach and similar vegetative crops appear to use food value, land and labor resources more efficiently than fruit crops such as watermelons, sweet corn and cucumbers. In the first group the original vegetative organs are used as food; in the second, the vegetative parts are not eaten directly, but produce an edible fruit. Thus our common foliage, stem, root and immature flower crops could be considered a "short chain"; our vegetable fruit crops a "long chain."

An extreme case of a long food chain has been cited by Kunkel¹ in connection with the food habits of the Eskimo. There are four organisms involved in this chain: the Eskimo lives largely upon seal meat, the seals eat fish, the fish consume snails and other invertebrates, and these in turn feed upon seaweeds. About 625 pounds of seaweed is required originally to produce a gain of 1 pound in the weight of the Eskimo. If the Eskimo would eat fish instead of seal meat, the chain would be shortened, and only 125 pounds of seaweed would be required. In pork production the data are expressed in different terms; but, as Hogan, Weaver, Edinger and Trowbridge² have pointed out, only about 40 per cent. of the energy consumed is stored in the animal's tissues. The same workers find that only about 10 per cent. of the protein consumed was stored in the tissues of the swine in this experiment. These values would vary somewhat with the age of the animals and with feeding conditions.

Foods for livestock may be classified into three groups on the basis of the competition between meatproducing animals and human beings. First, there are foods that may be used either by livestock or people, such as wheat and corn. Second, there are livestock feeds which are not usable by people, but which are grown upon land that could have produced human food; an example is alfalfa hay on irrigated

¹ B. W. Kunkel, Scientific Monthly, 46: 47-58, 1938. ² A. G. Hogan, L. A. Weaver, A. T. Edinger and E. A. Trowbridge, Mo. Agr. Expt. Sta. Res. Bul. 73, 1925.