of both sets of plants used in the present experiments (35 per cent of 7·9 = 2·76 thousand million in the 'high light' series as compared with 70 per cent of 3·9 = 2·76 thousand million in the 'low light' set). The plant, therefore, may be the major source of amino-acids for these organisms requiring them.

The significance of these observations with regard to plant nutrition, growth or susceptibility to disease is difficult to assess, especially since the role of the normal rhizosphere soil and root-surface microflora is still imperfectly understood. Eaton and Rigler[9] have reported decreased susceptibility of cotton plants to *Phymatotrichum omnivorum* as a result of defoliation or mutilation of the plants, which increased the carbohydrate content of the roots. This effect was associated also with a reduction in total bacterial numbers and an increase in numbers of fluorescent bacteria. Harley and Waid[10] observed an increase in the incidence of semi-pathogenic fungi on roots of beech seedlings grown in the shade. Whether these results are due to a concomitant reduction of the bacterial and possibly of the actinomycete population at the root surface, to an alteration of the types of organisms, as suggested above, or to changes in the plant and in the root which render them less able to withstand parasitic attack is not clear at present although the increased susceptibility of plants growing under reduced illumination to diseases produced by viruses, fungi and nematodes has been reasonably well established[11,12].

J. W. Rouatt
H. Katznelson

Microbiology Research Institute,
Research Branch,
Canada Department of Agriculture,
Ottawa.

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Iron Bacteria in Mine-waters

As van Beneden[1] has shown, iron bacteria may be obtained from fresh-water by reducing the iron in this water by percolation through a layer of soil. In the thin layer where the effluent oxidizes (presumably ferrous carbonate or bicarbonate to hydrated ferric oxide) the bacteria develop.

Baas Becking, Wood and Kaplan[2], using this method, obtained iron bacteria from sea-water. With the help of Mr. A. D. Haldane, senior chemist of the Bureau of Mineral Resources, a more efficient apparatus was constructed. Iron bacteria were grown on slides on which a reduced fresh-water effluent was dripping. Tap-water can be made free of oxygen by passing it through a column of steel-wool, previously washed in carbon tetrachloride. The effluent contains a slate-blue compound, possibly ferrous hydroxide, which oxidizes slowly to hydrated ferric oxide. Ferrous carbonate (and possibly the soluble bicarbonate) are formed and oxidized in culture flasks containing the infection material and a small amount of magnesium-ammonium phosphate.

As infection material, chalybeate waters from various localities in New South Wales were used (Matraville, Mittagong, Warrah, Palm Cave, Bungendore). Both a sheathed rod and a species of *Sphaerotilus* made their appearance. The method, however, is cumbersome and unsuitable for pure culture work.

The oxidation of pyrite proceeds in sterile suspensions, yielding ferrous sulphate and acid. In sterile suspensions, the oxidation of ferrous to ferric sulphate is very slow, only a few per cent being converted in four months at room temperature.

Iron bacteria isolated from chalybeate waters are capable of catalysing the oxidation of ferrous to ferric sulphate, the reaction running to completion in a few weeks at room temperature. It is possible to obtain pure cultures of these forms on 1 per cent ferrous sulphate agar containing 200 mgm./l. magnesium-ammonium phosphate. Pure cultures could also be obtained on powdered pyrite agar.

One of the final oxidation productions of pyrite is a mixture of ferric sulphates and oxysulphates, called 'yellowboy' by the miners. This ore, collected from various levels, Lake George Mine, Captains Flat, New South Wales, proved to contain *Sphaerotilus* and also *Gallionella*. The former could be isolated on 1 per cent ferrous-sulphate agar and on pyrite agar. Mine-water from the same mine contained *Sphaerotilus*.

A constant companion of both *Sphaerotilus* and the bacillary form is a small *Torulopsis*-like yeast, present in both mine-water and fresh-water, which also appeared from hydrated iron oxide collected near a hot spring at Talasea, New Britain.

Gallionella could not be isolated on sulphate media, but there are, apparently, iron organisms capable of developing on the ferrous (bi)carbonate as well as on the ferrous sulphate system.

In the latter system they give rise to electrode potentials of +860 mV.

The first stage in the oxidation of pyrite, where ferrous sulphate and acid are formed (*p*H 1·5–3·0), is a purely chemical phase, as the oxidation proceeds with equal intensity in sterile and in infected materials. In the second phase the biological catalyst will create an environment with very high electrode potentials, the *p*H remaining almost the same.

In old plate cultures of *Sphaerotilus* sp. a crust was formed around the bacterial colonies. In certain instances material from these crusts showed, in concentrated solution, a negative *p*H and potentials of more than +900 mV. There are more than 40 ferrous and ferric sulphate minerals described in the literature, and almost invariably they seem to be formed by the oxidation of pyrite in a dry atmosphere.

We can say, therefore, that the desiccation of the bacterial end-products adds a third, final stage to the oxidation of pyrite—a physical phase.

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L. G. M. Baas Becking
D. Moore

Bureau of Mineral Resources,
Department of National Development,
Turner, Canberra.

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