Review: Nitrogen-Fixation by *Azotobacter Chroococcum*
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into a northern and a southern cold water zone and a tropical zone between these. The tropical plankton was found to be poorer in number of organisms but richer in species than that of cooler waters. The Coccolithophoridae play an important part in all parts of the ocean, and the Peridineae are also widely distributed, while the Trichodesmieae (belonging to the family Oscillatoriaceae of blue-green algae) are exclusively tropical. On the other hand the Diatoms and the naked Phytoflagellata predominate in the cooler zones; the former are more numerous in the north, the latter in the south. In the zone three regions are distinguished both hydrographically and biologically—the northern region (Sargasso Sea and north equatorial current), the Guinea current, and the southern region (south equatorial current and Brazil current). The median portion of the Sargasso Sea showed a fair abundance of Coccolithophoridae, which fell off in numbers towards the margin of this sea and were found very sparingly in the Guinea current. In the latter were found abundant Trichodesmieae, also a *Euglena*, a *Prorocentrum* (Peridineae), and the Tunicate animal *Oikopleura dioica*—the last three organisms are apparently characteristic of this current, while *Trichodesmium* is also common in the north equatorial current but was not found in the south tropical region, where there were obtained enormous quantities of Coccolithophoridae belonging to species which were either new or had previously been found only sparingly. In the Brazil stream these organisms were accompanied by Diatoms, of which one (*Brenneckella Lorenzeni*) apparently lives in symbiosis with the Coccolithophorid *Pontosphaera sessilis*.

With regard to the vertical distribution of the plankton, in the cold water zones the maximum occurred near the surface, while in the tropics it was at about 50 m. depth; but a remarkable exception to this rule was found in the regions rich in *Trichodesmium* where this alga formed extensive floating masses at the surface. On the whole the ecological temperature optimum for the phytoplankton in the tropical zone lies in the deeper water; in the cold water zones the uppermost 100 m. contain 80—90 per cent. of this plankton, in the tropics less than 70 per cent. The author gives descriptions of a large number of new plankton forms, with special reference to their adaptations for flotation. Perhaps the most interesting result of the investigation—in which the most effective methods of centrifugal separation, etc., were employed—is that relating to the Coccolithophoridae; this group, the knowledge of which has grown rapidly during recent years, proves to have quite as varied and remarkable flotation mechanisms as those found in the Diatoms.

NITROGEN-FIXATION BY *AZOTOBACTER CHROOCOCCUM*

Mockeridge, Florence A. "Some conditions influencing the fixation of nitrogen by *Azotobacter* and the growth of the organism." *Annals of Botany*, **26**, 1912, pp. 871—887.

Since nitric acid is practically the only form in which higher plants obtain the nitrogen they require, the fertility of the soil is closely related to the maintenance by soil organisms of the nitrogen cycle of change, and these organisms have received a very large amount of attention. The organic nitrogenous compounds, largely proteins, are broken down by soil bacteria into simpler compounds, like the amino-acids, and these into ammonia which in turn is oxidised successively into nitrous and nitric acid by other organisms. Under certain conditions, still other organisms come into play and by their action the nitrogenous compounds are converted into free nitrogen gas and thus lost to the soil; while on the other hand another group of organisms cause the fixation of free nitrogen, combining it with oxygen, hydrogen, carbon, etc., into forms available for higher plants.

Among the nitrogen-fixing organisms, some, as is well known, live in symbiosis with green plants (Leguminosae, etc.), the bacteria obtaining the carbohydrate they must consume from the higher plant which is supplied by them with combined nitrogen. Of the nitrogen-fixing organisms living free in the soil, the most fully investigated are *Clostridium Pasteurianum* and *Azotobacter chroococcum*, both very widely diffused in the soil. The former is readily isolated from pond mud and other soils in which organic matter is decaying under more or less anaërobic conditions; but the extent of the nitrogen-fixation which it effects is relatively small—in the

laboratory not more than 2 to 3 mg. of nitrogen for each gram of carbohydrate consumed. *Azotobacter chroococcum*—a large oval organism 4 to 5 μ long and 3 μ broad, differing from most bacteria in containing glycogen and therefore staining deep brown with iodine—is aërobic, and a strong oxidising agent; the dextrose or other carbohydrate which it consumes is converted into carbon dioxide and water, with small quantities of lactic and acetic acids, alcohol, and sometimes (as in the case of *Clostridium Pasteurianum* also) butyric acid. *A. chroococcum* and allied forms are very widely distributed in soils in all parts of the world, having been found in most cultivated soils and also in many virgin soils—in short, it appears to be practically ubiquitous, though not occurring in “acid” soils. To *Azotobacter* and allied forms may be attributed an important part in preparing and maintaining the world’s stock of combined nitrogen; the richness in nitrogen of such virgin soils as the black soils of the Russian steppes, Manitoba, Argentina, etc., is probably due, not simply to the accumulation of vegetable matter (since plants other than Leguminosae do not themselves fix nitrogen), but to the action of *Azotobacter* which derives from this accumulating oxidisable material the energy required for fixation of nitrogen. The continued storing-up of nitrogen in virgin soils is made possible by the constant return to the soil of this oxidisable organic matter; this return forms the chief distinction between wild and cultivated land.

The authoress of the paper cited above, like other investigators of the biology of *Azotobacter*, had found considerable difficulty in obtaining and keeping pure liquid cultures of the organism, and had tried in turn most of the many different nutrient media and methods of culture recommended by various writers. The results were so unsatisfactory that she at last decided to make a thorough examination of the behaviour of *Azotobacter* in these different culture solutions, both in respect of the rapidity and purity of the growth and the extent of fixation of nitrogen obtained per unit of the carbohydrate consumed. The medium used fulfilled the requirements given by Mm. Krzemieniewski (*Bull. Acad. Sci. Cracovie*, 1910) as to the minimum amounts of the elements requisite for the consumption of 1 gram of dextrose—0.38 mg. potassium, 0.36 mg. calcium, 0.35 mg. magnesium, 2.46 mg. phosphorus, and 0.5 mg. sulphur. The sand-slope method of culture, by increasing the surface and therefore the supply of oxygen and nitrogen to the organisms, results in a more rapid, vigorous, and healthy growth, giving a much greater increase in nitrogen in a given time, and being much more free from contamination with anaërobic organisms like *Clostridium* (the growth of which is inhibited by aëration of the culture) than the method of ordinary liquid cultures. The use of the sand-slope method with a suitable culture medium results in the fixation of 14 mg. of nitrogen per gram of sugar (mannite) consumed, when the most vigorous organisms are used for purposes of inoculation. The presence in the medium of an excess of calcium or magnesium carbonate, as a neutralising agent, is more advantageous than that of sodium hydrate, and the former substances not only assist the rapidity of the growth but also help to maintain its purity. The presence of sodium salts is not only unnecessary but exerts a depressing influence on the activity of *Azotobacter* as regards the fixation of nitrogen; the beneficial effect of the sodium chloride solution in inoculating agar plate cultures is due to the fact that this liquid is isotonic with the cell-contents, a solution of similar concentration of many other salts answering the purpose equally well.

NITRIFICATION IN DIFFERENT SOILS

Fischer, H. “Versuche über Stickstoffumsetzungen in verschiedenen Böden.”
Landw. Jahrb., **41**, 1912, p. 755.

The author found that nitrification takes place more rapidly in a somewhat heavy soil than in a quite light sand. One of the causes is evidently the lack of lime in the latter soil. On the other hand, the formation of ammonia is greater in light than in heavy soils. The theoretical quantity of lime calculated as being necessary for the nitrification of a given quantity of ammonia does not in reality suffice for complete nitrification, this process attaining its maximum rapidity with about three times the theoretical amount of lime. Nitrification is favoured by the supply of organic materials such as sugar or peat-decoction, but when too much nitrogenous material is