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# Rothamsted Report for 1932

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## Microbiology

### Rothamsted Research

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## MICROBIOLOGY

(Departments of Bacteriology, Fermentation and General  
Microbiology)

### (a) BACTERIA

- XLVI. E. MCCOY. "Infection by Bact. Radicicla in relation to the Microchemistry of the Host's Cell Walls." Proceedings of the Royal Society, B, 1932. Vol. CX, pp. 514-533.

It is statistically proved that infection of the root hairs is not a mere invasion of mechanically injured or broken root hairs. The presence of the bacteria, even of strains belonging to foreign inoculation-groups, causes a significant increase in the number of curled and bent hairs. The bacteria produce a secretion capable of modifying the wall, as evidenced by the abnormal curling of the root-hair tips. This secretion is separable from the cells by filtration, and is not specific for the plants of the cross-inoculation group to which the bacteria belong.

The bacteria in culture were unable to attack cellulose, pectin or calcium pectate. Curled tips of root hairs, whether infected or not, contain the same constituents as normal hairs. These constituents are cellulose, calcium pectate, and probably pectose, and a very resistant hemicellulose.

The cell walls of the nodule contain cellulose, a hemicellulose, calcium pectate in the mature parts, and pectose at least in the meristematic tip. Walls of the tip also give a protein reaction. There are numerous pits perforating the secondary layers of the walls, but the middle lamellae appear to be continuous. These pits are of sufficient size to admit infection threads, and it is suggested that the bacterial zoogloea crosses a cell wall by way of the pits. The infection thread is surrounded by a definite sheath consisting of cellulose and hemicellulose; calcium pectate is absent and the presence of other pectic materials has not been confirmed. The sheath is probably a deposit of the individual plant-cell.

- XLVII. JADWIGA ZIEMIECKA. "The Azotobacter Test of Soil Fertility applied to the Classical Fields at Rothamsted." Journal of Agricultural Science, 1932, Vol. XXII, pp. 797-810.

The kneaded plate (*plaque moulée*) method of detecting deficiency in lime and available phosphate was applied to 79 soil samples taken from the classical Rothamsted arable plots.

The test correctly indicated whether phosphate had been applied in soils receiving little or no nitrogen manures. In soils receiving 86 lb. or more mineral nitrogen per acre, the test usually showed little or no *Azotobacter* growth, even in the presence of phosphate and calcium carbonate. Silica jelly counts showed that *Azotobacter* cells were very much reduced in number in such soils. In some cases the test was modified by inoculating the sample with a culture of *Azotobacter* and it then gave correct indications as to phosphate content.

In general, *Azotobacter*, when present, was found to develop on kneaded plates, if the soil contained at least 10 mg. of water-soluble  $P_2O_5$  per kilogram of soil, but below this limit little growth occurred.



(b) PROTOZOA

- XLVIII. D. WARD CUTLER, L. M. CRUMP AND A. DIXON. "Some Factors Influencing the Distribution of Certain Protozoa in Biological Filters." *Journal of Animal Ecology*, 1932, Vol. I, pp. 143-151.

The purity of a medium, as measured by the amount of reducing material present in the solution, and the food supply, are two of the principal factors influencing the distribution of protozoa in sewage filters. The protozoa considered occur throughout a wide range of pH values, but the optima for different species are different.

Where chemical compounds added to the solution affect the protozoan population adversely, it may be due either to the formation of deleterious oxidation products, or to the development of a bacterial flora which is inimical to the protozoa.

(c) BIOLOGICAL ACTIVITIES

- XLIX. (a) A. G. NORMAN. "The Biological Decomposition of Plant Materials. VII. The nature of the residual hemicelluloses of rotted straw." *Biochemical Journal*, 1932, Vol. XXVI, pp. 573-577.

The nature of the residual hemicelluloses of well-rotted straw has been investigated. Only very small quantities were obtained, and there was no indication of variation in availability or the accumulation of less available groupings. The results indicate that the distribution and arrangement of the hemicelluloses in the cell-wall are such that microbial attack is not hindered by the presence of any resistant barrier. A water-soluble polysaccharide, probably of microbial origin, was also prepared. It contained 33 per cent uronic acid anhydride, and 66 per cent hexosan, and gave evidence of the presence of glucose units.

- XLIX. (b) A. G. NORMAN. "The Biological Decomposition of Plant Materials. VIII. The Availability of the Nitrogen of Fungal Tissues." *Annals of Applied Biology*, 1933. Vol. XX, pp. 146-164.

Fungal tissue was found to be as suitable a source of nitrogen as ammonium salts or nitrates for the decomposition of straw both by mixed soil flora and by pure cultures of certain fungi. Nitrification in soils of a number of samples of fungus tissue was compared with that of artificial mixtures of equal C/N ratio built up from glucose, cellulose and straw, each with added inorganic nitrogen. A clear correlation was found between the C/N ratio of the fungal material and the nitrogen nitrified. In all cases fungus tissue was at least as readily nitrified as the artificial mixtures. No evidence was found for the existence of a very resistant and unnitrifiable residue from fungus tissue.

- L. S. H. JENKINS. "The Biological Oxidation of Carbohydrate Solutions. II. The Oxidation of Sucrose in the Presence of Different Inorganic Nitrogen Compounds." *Biochemical Journal*, 1933, Vol. XXVII, pp. 245-257.

The effect of different sources of nitrogen on the biological oxidation of sucrose through a percolating filter was studied. Under



the conditions of filtration in these experiments there was a considerable disappearance of nitrogen from solutions having C/N ratios of 8.4/1 and 4.2/1, irrespective of the form in which the nitrogen was supplied. Greatest disappearance of nitrogen occurred with nitrite and less with ammonia. The apparent losses when nitrogen was supplied as nitrite and nitrate were most marked in that part of the filter in which carbohydrate oxidation was most active. These apparent losses include the nitrogen immobilised by the micro-organisms of the film, and it was impossible to state how much of this was due to losses of elementary nitrogen.

- LI. S. H. JENKINS. "*The Biological Oxidation of Carbohydrate Solutions. III. Nitrogen, Phosphorus and Potassium Balances in Percolating Filters.*" *Biochemical Journal*, 1933, Vol. XXVII, pp. 258-273.

By studying the decomposition of sucrose in percolating filters filled with glass, and so allowing the recovery of the synthesised film, it was possible to draw up balance sheets for the nitrogen, phosphorus and potassium salts added. The recovery of the last two elements was not quantitative, possibly owing to inadequate methods of analysis. In experiments containing sugar and ammonium salts, giving a C/N ratio of 8.4/1, the balance sheets for nitrogen showed that about 14 per cent. of that supplied was lost. With a ratio of 8.4/1 a slight gain was recorded. When filters were supplied with an ammonium salt as the source of nitrogen neither nitrite nor nitrate was detected in the effluents. When the source of nitrogen supplied to the filters was organic neither ammonia nor oxidised compounds of nitrogen were found. The observed losses, therefore, could not have taken place through formation of ammonia or the production of nitrite or nitrate and subsequent denitrification. Liberation of elementary nitrogen is probably carried out entirely within the cells of the organisms.

- LII. S. H. JENKINS. "*The Design of Experimental Percolating Filters.*" *Biochemical Journal*, 1933, Vol. XXVII, pp. 240-244.

Percolating filters of a new design have been constructed composed of sections which fit together so that no air spaces occur between individual sections. These have been made in wood built from six octagonal units, the top and bottom edges of each being bevelled at an angle of 60°. The medium was supported in each section by means of a rustless steel tray. Similar filters have been made with cylindrical glass units joined together by means of wide bands of rubber. The medium was held in each section on a perforated aluminium plate cemented to the bottom of each cylinder. Such filters may be operated as a whole or the changes taking place at any given depth investigated by sampling.

- LIII. J. MEIKLEJOHN. "*The Effect of Colpidium on Ammonia Production by Soil Bacteria.*" *Annals of Applied Biology*, 1932, Vol. XIX, pp. 584-608.

In two series of experiments using different media (peptone dissolved in soil extract in the first series, and a synthetic medium



containing alanine in the second), cultures containing the ciliate protozoon *Colpidium* with two species of soil bacteria were compared against control cultures containing only the two species of bacteria.

On both media an appreciable reduction in bacterial numbers, as compared with the numbers in the control cultures, was observed in the *Colpidium* cultures, but in spite of this reduction, the *Colpidium* cultures produced more ammonia from peptone than the controls, and nearly the same amount of ammonia and carbon dioxide from alanine as the controls.

In both series of experiments an inverse linear relation was found to exist between total bacterial numbers and the amount of ammonia or carbon dioxide produced per 1,000 million bacteria (efficiency).

In the second series of experiments, the regression coefficients of efficiency on average bacterial numbers are significantly different in the *Colpidium* and the control cultures.

It follows that the presence of *Colpidium* has a stimulating effect on ammonia production, which is not due solely to the reduction of bacterial numbers to an optimum value, and it is suggested that in the cultures in which *Colpidia* are present, the bacteria are kept in a state of physiological youth for a longer period than the normal.

LIV. D. WARD CUTLER AND L. M. CRUMP. "*Some Aspects of the Physiology of Certain Nitrite-Forming Bacteria.*" *Annals of Applied Biology*, 1933, Vol. XX, pp. 291-296.

One hundred and four species of bacteria which produce small quantities of nitrite from ammonium sulphate have been isolated from filters receiving waste water from a beet sugar factory and these bacteria do not differ in their behaviour on carbohydrates from non-nitrifying bacteria obtained from the same source.

Ammonium lactate is more readily oxidised than is ammonium carbonate, phosphate, sulphate or acetate, and in the majority of cases nitrite itself can also be utilised by these bacteria in the course of growth.

There is positive correlation between increase in bacteria numbers and the percentage nitrite in a culture during the initial growth period and there is evidence that nitrite may disappear slowly from acid solutions without the intervention of bacteria, though this is not invariably the case.

LV. N. W. BARRITT. "*The Nitrification Process in Soils and Biological Filters.*" *Annals of Applied Biology*, 1933, Vol. XX, pp. 165-184.

Nitrifying cultures in mineral salt solutions were obtained from laboratory percolating filters and resembled similar cultures obtained from soils in their ability to grow on silica gel plates and in showing a low thermal death point. The addition of organic matter depressed the rate of nitrification in comparison with the rate of nitrification in the presence of mineral carbonates. The addition of a solution of carbonic acid also depressed nitrification, and it is inferred that the supposed toxic effect of organic matter is not a direct one, but due to the liberation of excessive amounts of CO<sub>2</sub>.

The optimum pH for nitrification is between 6.7 and 8.0. It ceases at pH 9.2 and 5.5, at which point the free acid is spontaneously oxidised to nitric acid without the aid of a specific organism.



An increase in nitrifying power of soil after passage through earthworms is recorded and accounted for by the digestion of organic matter and addition of  $\text{CaCO}_3$  from the subsoil.

It is suggested that the evidence of many workers points to a possible autotrophic phase in the life cycle of heterotrophic organisms.

## THE PLANT IN DISEASE : CONTROL OF DISEASE

(Departments of Entomology, Insecticides and Fungicides, and Plant Pathology)

### (a) INSECTS AND THEIR CONTROL

- LVI. H. F. BARNES. "*Studies of Fluctuations in Insect Populations. I. The Infestation of Broadbalk Wheat by the Wheat Blossom Midges (Cecidomyiidae).*" *Journal of Animal Ecology*, 1932, Vol. I, pp. 12-31.

Fluctuations of insect populations are being studied in three directions : (1) the intensity of attack by the larvae ; (2) the degree of parasitism ; and (3) the dates of emergence and number of broods. Study of the two wheat blossom midges reveals considerable fluctuations in intensity of attack and the extent to which they are parasitised by other insects. Extensive new information regarding the bionomics of these two midges, *C. tritici* and *S. mosellana*, is given.

- LVII. H. F. BARNES. "*A Study of the Segmentation of the Antennae in Gall Midges.*" *Proceedings of the Zoological Society of London*, 1932, pp. 323-334.

From a study of over 14,300 individuals of fourteen species of economic importance, it is shown that, in some species and genera, food affects the size of the adult midges only ; in others it affects the size of adult midges and, in addition, the number of antennal segments. A formula is given for the frequency and range in the number of antennal segments.

- LVIII. H. F. BARNES. "*On the Gall Midges Injurious to the Cultivation of Willows. I. The Bat Willow Gall Midge (Rhabdophaga terminalis H.Lw.)*" *Annals of Applied Biology*, 1932, Vol. XIX, pp. 243-252.

The bionomics of the bat willow gall midge, which does serious damage to certain willows grown for basket-making and the cricket bat willow grown for sets, are described. The midge exhibits a distinct host-plant preference, choosing the bat willow (*S. coerulea*) when possible. But it also breeds readily on a golden willow (*S. alba* var. *vitellina*). It will not attack Black Maul (*S. triandra*), Long Skin (*S. viminalis*) and Dicky Meadow (*S. purpurea*).

- LIX. MARGOT E. METCALFE. "*Dasyneura leguminicola (Lint.), the Clover Seed Midge.*" *Annals of Applied Biology*, 1933, Vol. XX, pp. 185-204.

An attempt was made, after studying the biology of this midge, to find resistant or immune varieties of red clover. It is suggested that clovers grown for seed production should be in the green-head