a concentration of 0·025 per cent of acetylcholine, the increase in enzyme activity by calcium and magnesium amounts to 100–150 per cent, the inhibition by potassium to about 40 per cent. The inhibition by potassium ions is abolished by calcium or magnesium. In a balanced medium, such as Tyrode solution, a decrease in calcium or magnesium has the same effect on esterase activity as an increase in potassium ions. All these effects are brought about by physiological concentrations of the electrolytes. The following table illustrates these facts.

HYDROLYSIS OF ACETYLCHOLINE.

0·05 c.c. purified cholinesterase preparation from horse serum; 0·5 mgm. acetylcholine; 2 c.c. 0·025 mol. sodium bicarbonate; 5 per cent carbon dioxide in nitrogen; 37° C.

Cations added (in form of chlorides).	Acetylcholine hydrolysed in 3 minutes
—	124 γ
0·24 mgm.% Mg	174 γ
2·4 „ Mg	249 γ
0·44 „ Ca	180 γ
2·2 „ Ca	288 γ
400 „ K	73 γ
400 „ K + 0·72 mgm.% Mg	129 γ
400 „ K + 0·55 „ Ca	133 γ

The adjuvant actions of potassium on parasympathetic effects and the depressant actions of calcium (and magnesium) on such effects may be explained by the effects of these electrolytes on cholinesterase. Potassium, by inhibiting the esterase activity, would delay the destruction of acetylcholine and thus augment parasympathomimetic effects; calcium and magnesium, on the other hand, by activating the esterase, would accelerate the hydrolysis of acetylcholine, and thus antagonize such effects.

In the same way, the hyper-irritability in the various forms of tetany, known to be associated with a low calcium or magnesium level, may possibly be due to a delayed destruction of the acetylcholine liberated by a motor nerve impulse at the nerve endings. The vasodilatation, observed in magnesium deficiency, may similarly be due to delayed destruction of acetylcholine liberated by impulses in cholinergic vasodilator nerves.

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Can Rusts Fix Nitrogen?

Caldwell *et al.*[1] and Murphy[2] have shown that in wheat and oat plants infected with *Puccinia triticina* and *P. coronata* respectively, the amount of nitrogen, and of some other substances present, is greater than in uninfected plants. Gretschushnikoff[3] found that the rust he studied excreted urea and ammonia, which, he suggested, were removed by the host, since no correlation was found between the severity of the infection and the amount of ammonia and urea in the extracts of the rusted leaves. These facts suggest that rusts may fix atmospheric nitrogen just as some other fungi are believed to do.

Work begun in Cambridge in 1937 and continued in Portugal has suggested that the problem is worth investigating in some detail. The results of two experiments are given below. Six samples each of fifty plants of Jenkin wheat were grown on sand until the plants were showing the second leaf, and after cutting off the roots and grain the plants were washed in distilled water. Samples 1 and 2 were taken at the beginning of the experiment; samples 3 and 4 were placed with their cut ends in water and kept rust free; samples 5 and 6 were placed in water and inoculated with *P. graminis tritici*, race 27. Sample 4 was accidentally destroyed before the close of the experiment. Samples 3, 5 and 6 were collected twelve days after the experiment was begun. The plants when collected were dried and then analysed. The results of the analyses are given in the accompanying table.

	Rust-free	Dry weight in gm.	Absolute nitrogen per gm.
Initial control	1	1·2549	0·06804
Final control	2	1·2858	0·06038
	3	1·6790	0·06230
	Inoculated		
	5	1·5615	0·07335
	6	1·4406	0·07980

In the second experiment, seedlings of Spratt Archer barley were used. The seedlings were prepared in the way described above; fifty plants were dried for analysis when the experiment was begun. Two samples of 225 plants each were placed with their cut ends in modified Crone's nitrogen-free solution. One sample was kept rust free, the other was infected with *Puccinia anomala* race 12. Fifty seedlings were taken every three days from each sample and dried at once. After twelve days the experiment was discontinued and the samples analysed. The results of the analyses are given below:

		Dry weight in gm.	Absolute nitrogen in gm.
	Initial control	0·7206	0·03012
	Rust-free		
No. of days the seedlings were kept in the solution	3	0·8853	0·0326
	6	0·9908	0·03073
	9	1·0067	0·0326
	12	1·8007	0·02996
	Inoculated		
No. of days the seedlings were kept in the solution	3	0·9156	0·033015
	6	1·1150	0·03458
	9	0·9922	0·03542
	12	0·8836	0·01809

The results of other experiments not given here agree in essentials with those quoted above, although in some experiments with *P. glumarum* the apparent increase in nitrogen content was not so marked. Irregular results were found with other rusts, particularly if the leaves remained wilted for long periods after inoculation. Nevertheless the experiments point to an increase in the amount of nitrogen in the rust-infected plants.

I realize the difficulties involved in demonstrating nitrogen fixation by an obligate parasite, yet I feel that the above results are sufficiently suggestive to warrant their being brought to the notice of other workers.

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[1] Caldwell, Craybill, Sullivan and Compton, *J. Agric. Res.*, **48**, 1049–1071 (1934).
[2] Murphy, H. C., *Phytopath.*, **26**, 220–234 (1936).
[3] Gretschushnikoff, A. I., *C.R. Acad. Sci. U.R.S.S.*, N.S., **2**, 335–340 (1936); (Ref. in *Rev. Appl. Mycol.*, **15**, 710; 1936).