

unrelated, antibiotics. The development of this multiple resistance occurred in a gradual stepwise manner, and there was no evidence that polyresistant variants were present in the original antibiotic-sensitive cultures.

In contrast to these findings, the multiple-resistant variants described here were, in general, as sensitive to streptomycin as was the parent strain, and streptomycin-resistant variants of the original culture of *Salm. paratyphi B* (B.R.L. 1824) remained sensitive to the penicillins, chloramphenicol and tetracycline. Likewise, a culture of this organism repeatedly transferred in the presence of ampicillin showed no change in sensitivity to cycloserine, kanamycin, neomycin or polymyxin.

The development of cross-resistance involving penicillins, chloramphenicol and tetracycline was also demonstrated with a strain of *Escherichia coli*, a strain of *Klebsiella pneumoniae* and several *Salmonella* species. Cross-resistance was not seen with two strains of *Proteus mirabilis*, and one strain each of *Shigella flexneri* and *Shigella sonnei*.

It would seem, therefore, on the basis of the findings reported here, that the emergence of antibiotic-resistance *in vitro* should not be regarded as a phenomenon which is highly specific for the antibiotic in which the bacterial culture is transferred. On the contrary, it is evident that cross-resistance among dissimilar antibiotics can readily be demonstrated *in vitro* with certain bacteria, particularly species of *Salmonella*, although the clinical significance of this is as yet not known.

These results will be reported in detail elsewhere.

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Role of Cobalt in Nitrogen Fixation by *Azotobacter chroococcum*

COBALT is known to accelerate nitrogen fixation by the blue-green algae, and the lowest cobalt requirement of *Nostoc muscorum* was observed to be 0.05 p.p.m.¹ Its influence on *Azotobacter* does not seem to have been examined critically. The experiment recorded here was therefore planned to examine the effect of cobalt at different concentrations varying from 0.02 to 5.0 p.p.m. on nitrogen fixation by *Azotobacter chroococcum* in Jensen's medium². The results obtained were compared with those observed with molybdenum, which was already known to be essential for *Azotobacter*. The basal medium was tested for cobalt and molybdenum by 'Nitroso-R-salt' method³ and *Aspergillus niger* method respectively. While cobalt could not be detected in the medium, molybdenum of the order of 0.00028 p.p.m. was found to be present. The cobalt salt $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ used was also tested for molybdenum by the same method as mentioned here, and it contained 0.0001 p.p.m. of molybdenum at 0.1 p.p.m. concentration of cobalt, at which nitrogen fixation was found to be enhanced.

The results indicated that cobalt at a dose as small as 0.1 p.p.m. increased the amount of nitrogen fixed. At 1.0 p.p.m. and higher concentrations, however, the response was less. Molybdenum was found to accelerate nitrogen fixation at and above a concentration of 1.0 p.p.m. examined. Higher concentration of molybdenum up

Table 1. EFFECT OF COBALT AND MOLYBDENUM IN NITROGEN FIXATION BY *Azotobacter chroococcum* IN JENSEN'S MEDIUM

Conc. (p.p.m.)	Nitrogen fixed in mg/g of sucrose oxidized (average of 4 replications)		
	Molybdenum	Cobalt	
0.00	6.06	6.06	
0.02	6.47	6.22	
0.10	6.35	9.00	
1.00	13.10	8.35	
2.00	13.82	8.22	
3.00	13.25	8.20	
4.00	13.25	7.90	
5.00	13.88	7.95	
S.E.	0.069	S.E.	0.090
C.D. at 5% level for Mo	0.21	C.D. at 5% level for Co	0.27

to 5.0 p.p.m. examined did not result in any further increase in this value.

In order to find out the extent to which molybdenum as impurity in the cobalt salt would influence nitrogen fixation, another experiment was conducted with a control, 0.0001 p.p.m. of molybdenum and cobalt (0.1 p.p.m.), before and after removal of traces of molybdenum from the culture medium following the method of Nicholas⁴.

Table 2. NITROGEN FIXED IN MG/G OF SUCROSE OXIDIZED (average of 4 replications)

	Before removal of Mo		After removal of Mo	
	Mo p.p.m.	Co p.p.m.	Mo p.p.m.	Co p.p.m.
Control	0.0001	0.1	0.0001	0.1
6.74	6.9	9.66	6.83	9.73
S.E.	0.3276		0.2148	
C.D. at 5% level	1.05		0.69	

From the foregoing it can be seen that enhanced nitrogen fixation observed was only due to cobalt and not to molybdenum present as impurity in traces.

Cobalt, being the centre of a complex in vitamin B₁₂, is likely to play an important part in plant and animal metabolism as vitamin B₁₂ is known to have a definite role in cell synthesis.

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Deoxyribonuclease Activity from 'Lactic Acid Pleuropneumonia-like Organisms'

THE relationship of the micro-organisms of the pleuropneumonia group (PPLO, Mycoplasmatales) to the true bacteria is uncertain. Two general explanations have been advanced to explain the nature of this group and its relation to other micro-organisms¹: one holds that pleuropneumonia organisms are a natural biological class; the other that PPLO are an assemblage of the stable *L* forms of various bacteria.

Previously we have emphasized the basic biochemical heterogeneity of the organisms making up the group² and proposed that primarily these organisms could be divided into sets consisting of fermentative and non-fermentative strains. These fermentative strains were characterized as catalase negative, nitrate negative aerobes that accumulate lactic acid. This description is highly reminiscent of certain lactic acid bacteria, particularly the *Streptococci*. I would term this set of organisms 'lactic acid pleuropneumonia-like organisms'.

Either the resemblance between this set of pleuropneumonia organisms and the lactic acid bacteria occurs because these *Mycoplasma* and the *Lactobacillaceae* are