yellowish-brown stain on a very pale background with phloroglucinol spray.

It has been found that for a spot-content of more than 100–250 μgm. of sucrose (depending upon the conditions mentioned later), the linear relation between spot-length and logarithm of spot-content breaks down, and a much truer representation is found by the logarithm of spot-content bearing a linear relation to the *logarithm* of spot-length. This has been tested and found to hold true over a range 4–450 μgm. of sucrose. It may be noted that at low concentrations of sugar, either the logarithm of spot-length or spot-length itself will tend to give a linear relation with the logarithm of spot-content.

In addition, an investigation of the factors determining the length of stain has been attempted, and there are indications that increase of temperature decreases absolute stain-length while increase in the time of running of the chromatogram has the opposite effect. Increase of the time allowed for equilibrium to be reached before running the chromatogram also increases absolute stain-length. For all the combinations of controlled temperature (35–40° C.), length of run, and time of equilibrium so far tested, the graphs of logarithm of stain-length against logarithm of spot-content have been parallel within the limits of experimental error. On the subject of slow attainment of equilibrium conditions, the recent paper of Müller and Clegg[4] can be consulted.

This work has been carried out as part of the programme in plant physiology of this Section. It is intended to publish a fuller account elsewhere.

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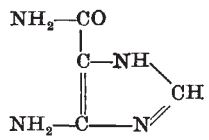
Ribosidation of 4-Amino-imidazole-5-carboxamide by *Escherichia coli*

It has already been shown[1,2] that 4-amino-imidazole-5-carboxamide (I) is a precursor of the purines in *E. coli*; a purine-requiring mutant of the organism grows in a medium containing this substance. On the other hand, it has been observed[3,4] that synthetic 4-amino-imidazole-5-carboxamide is little utilized by a growing culture of *E. coli*. The following observations will eliminate this apparent discrepancy.



(I)

(1) When a cell suspension of *E. coli B* is incubated with the hydrochloride of (I) in *M*/15 phosphate buffer (*p*H 7), and the supernatant is added to the purine-requiring *E. coli* mutant, a growth-enhancing effect is observed which is five times as great as that of the hydrochloride of (I).

(2) The supernatant containing the new factor was lyophilized and the concentrate subjected to paper chromatography according to Carter[5] in 5 per cent potassium dihydrogen phosphate – *iso*amyl alcohol and developed by diazotization. Incidentally, the presence of the original amino group in the new factor was thus established. While the hydrochloride of (I) has R_F 0·68, a new spot of R_F 0·63 was observed. The activity of the new spot was established by the bio-autographic technique of Winsten and Eigen[6].

(3) The fraction containing the new substance showed a positive orcin test and a colour reaction with diphenylamine[7] which exceeded that of the blank, indicating the presence of deoxyribose. The ultra-violet spectrum showed a shifted absorption maximum (2600 A.) as compared with the hydrochloride of (I) (2680 A.).

It seems thus established that 4-amino-imidazole-5-carboxamide is converted by *E. coli* into a glycoside.

(4) It has not been finally established whether the new substance is a nucleoside or a nucleotide. According to Carter[5], nucleotides cannot be chromatographed by means of butanol. Since the new spot could also be obtained in butanol, it seems likely that the new substance is a nucleoside (deoxyriboside of (I)). A similar substance may have been encountered by Friedman and Gots[8] in their recently reported experiments.

(5) In connexion with the mechanism of the conversion of (I) into purine bases, the following observation appears to be of interest. When (I) is added to a cell suspension of *E. coli* in phosphate buffer (*p*H 7) containing 0·2 per cent glucose, the diazotizable amino group disappears, and a substance is formed which has the activity of a nucleoside (it is eight times as active a growth factor for the purine-mutant as the hydrochloride of (I)). It appears that the catabolism of glucose can supply to the glycoside of (I) the C_2 atom of the purine system[3,9].

The question has often been asked[10] whether (I) is utilized as such or in the form of a glycoside. From the observation made here, and those reported recently by Greenberg[11] and by Schulman and Buchanan[12] for pigeon's liver preparations, it seems that the purine bases are formed via their ribosides and ribotides, and that in (I) the pyrimidine ring is closed only after ribosidation has occurred. According to S. E. Kerr *et al.*[13], however, adenine and guanine are incorporated by rapidly growing yeast into ribonucleic acid much *better* than the corresponding nucleosides and nucleotides.

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