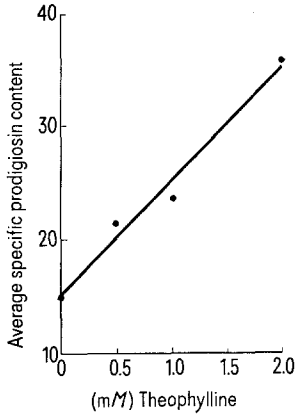
Discussion. It is postulated that the reversal of the glucose repression of prodigiosin synthesis by theophylline is the result of the inhibition of PDE and the concomitant increase in cellular cAMP concentration. However, further studies, involving the measurement of cellular cAMP levels, are required to substantiate the validity of this suggestion.

With reference to the studies of β-galactosidase induction and repression in catabolite repression sensitive and insensitive strains of *E. coli*[5], and their cAMP degradative abilities[2,6] it was not expected to achieve complete reversal of glucose repression of prodigiosin production and this was not achieved with the doses of theophylline utilized. However, 2 m*M* theophylline did cause approximately a doubling of specific content.



The concentration of theophylline plotted against the average specific prodigiosin content for glucose repressed cells of *S. marcescens*.

It is interesting to note that the reversal of glucose repressed motility in *E. coli* by theophylline has been reported[7], which ,if one assumes its action to be via PDE inhibition, is at variance with the in vitro *E. coli* inhibition study. Un reported work of mine failed to demonstrate theophylline reversal of glucose repressed β-galactosidase induction in *E. coli* K12, but this, however, could be due to the insensitivity of the enzyme, or to the inherent differing sensitivities of the different systems to the modulation of cAMP levels.

It is suggested that this work may form the basis for the development of a correlative screen for potential pharmacologically active compounds, the mode of action of which are, in part, or in total, the inhibition of PDE.

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Identification of Uncommon Amino Acids in the Lentil Seed (*Lens culinaris Med.*)

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Summary. The fraction of the free basic amino acids in the lentil seed was shown to contain γ-hydroxyarginine, γ-hydroxyornithine and homoarginine besides the common amino acids. Similar distribution was found in 5 varieties of lentil, with hydroxyarginine and arginine dominating. The significance of these findings with regard to chemotaxonomy and lentil consumption is discussed.

In a previous report, γ-hydroxyarginine and γ-hydroxyornithine were shown to occur as free amino acids in 5 varieties of lentil seeds[1]. These compounds were identified by comparison with synthetic samples on an amino acid analyser, under different elution conditions. As the native compounds from lentil are stereo-chemically uniform, and since chemical synthesis of each of these hydroxylated amino acids yields a mixture of the two possible diastereomeric forms, full elucidation of their structures could not be accomplished[2]. For further study, γ-hydroxyarginine, representing up to 70% of the total free basic amino acids in lentil seeds, was isolated from a seed extract by ion exchange chromatography[3]. An unidentified compound, showing a ninhydrine-positive reaction and eluted after arginine, was further isolated by the same separation procedure[4].

Treatment by 6 *N* hydrochloric acid, under reflux, left the compound unchanged. Boiling for 1 h with 2 *N* sodium hydroxide decomposed the sample to lysine, urea and decomposition products of the latter, in equimolar ratio. The presumption that it was homoarginine could be made by comparison with an authentic sample of homoarginine (mixed samples, using buffers of different pH values in the analyser).

The Table shows the distribution of the free basic amino acids in 5 varieties of lentils investigated[5]. Hydroxyarginine and arginine dominate over hydroxyornithine and the common basic amino acids. On the basis of these findings, homoarginine, as well as hydroxyarginine and hydroxyornithine, has to be considered a regular and specific component of the lentil seed. The occurrence of uncommon metabolites may be helpful to botanists for classification work, or for the study of phylogenetic relationships. The plant family of Leguminosae offers a great field of chemotaxonomic study. It should be men-

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[4] For experimental details concerning ion exchange chromatography and analysis of fractions, see ref.[3]. Homoarginine was collected between 10 and 12 l of eluant.

[5] The determination of the hydroxy amino acids is sensitive to variation, since these compounds are partially converted to γ-lactones, even under slightly acidic conditions. The concentrations given in the Table have, therefore, to be regarded as approximate values.

Occurrence of free basic amino acids in 5 varieties of lentil seed

Variety[a]	I	II	III	IV	V
Region	Balkans	USA	Algeria	India	France
Total nitrogen	4.10	4.49	4.00	4.26	3.80
Lysine	0.05	0.06	0.05	0.06	0.07
Histidine	0.02	0.02	0.01	0.03	0.01
Arginine	0.29	0.41	0.25	0.23	0.26
Ornithine[b]	n.d.	n.d.	n.d.	n.d.	n.d.
γ-Hydroxyornithine	0.06	0.08	0.04	0.06	0.06
γ-Hydroxyarginine	0.81	0.85	0.51	0.56	0.89
Homoarginine	0.02	0.07	0.02	0.02	0.01

All concentrations in % of dry substance. [a] For characterization of variety see reference[1]. [b] Present in small concentration; n.d. = not determined.

tioned that γ-hydroxyarginine and γ-hydroxyornithine were fond in several species of vetch (genus *Vicia*), and homoarginine, as a possible precursor of lathyrine, occurs in some species of the genus *Lathyrus*[6]. The presence in the lentil seed of both hydroxyarginine and homoarginine might be an incentive towards a review of the taxonomic position of the genus *Lens*.

Legumes constitute an important source of dietary protein for a considerable percentage of the world population. Certain peas of the genera *Lathyrus* and *Vicia*, which are of no commercial significance but which are eaten by sections of the population of India and the Mediterranean area, may cause neurological or muscular conditions, called lathyrism[7]. This disease is associated with the presence of toxic amino acids in these legume seeds, e.g. β-cyano-L-alanine, α,γ-diaminobutyric acid, β-N-oxalyl-α, β-diaminopropionic acid, and the related β-aminopropionitrile.

Lentil contains none of these toxic factors. Its characteristic and dominating amino acid within the legume family is γ-hydroxyarginine. Lentils have been eaten since ancient times, and have played an important role as a nutritional base in certain cultures of antiquity. This food has been proved 'historically' to be wholesome, yet its value has always been judged ambiguously. It is well known that some people suffer from vomiting, or allergic symptoms, after eating lentils. However, no scientific base, or experimental data, exists for considering a compound like hydroxyarginine as harmful, or as responsible for the phenomena mentioned. In preliminary inhibition studies with arginine antagonists, it was shown that γ-hydroxyarginine did not inhibit the growth of *Escherichia coli* or *Pseudomonas aeruginosa*[8]. Of lesser interest are similar experiments with homoarginine, which is a minor compound in the lentil seed[9,10].

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Assimilation of Ammonia and Growth of Biotin Deficient *Aspergillus nidulans*

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Summary. Biotin deficiency in *Aspergillus nidulans* has been found to increase the uptake of ammonium ions, associated with a marked increase in the activity of NADP-linked glutamate dehydrogenase, which is found to be the major route of ammonia assimilation in this culture. The results obtained are discussed with respect to the growth of *Aspergillus nidulans* during biotin deficiency.

Earlier studies in our laboratory indicated that biotin deficiency in *Aspergillus nidulans* gives rise to increased cellular synthesis, when grown on NH_4NO_3 as a sole nitrogen source[3,4]. Furthermore, it was observed that biotin deficiency in this culture caused marked increase in the growth rate, while the total period required for the completion of the growth cycle remained unaltered[5]. We have been able to demonstrate the remarkable increase in the uptake rate of ammonia attributable to biotin deficiency in *A. nidulans*[5,6]. Since biotin deficiency in this culture causes a marked increase (70%) in the protein content with a concomitant fall (65%) in the fatty acid content of the mold, it was of interest to study the assimilation of ammonia by this culture under these conditions. The present investigation suggests that the major route of ammonia assimilation is through NADP-linked glutamate dehydrogenase. The results obtained are discussed with respect to the growth of *A. nidulans* in a state of biotin deficiency.

The strain, media composition and the culture conditions used in the present investigation were the same as described earlier[5,7]. Culture grown in the presence of 5 units of avidin (General Biochemicals, Ohio) has a 65% lower fatty acid content than control. Biotin was not traceable in the mycelial extract, using the microbiological assay with *Lactobacillus arabinosus*, according to the

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