

Tryptophane Requirement of the Chick.

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In studies on the amino acid requirements of chicks it has been found necessary to ascertain the quantitative tryptophane requirement as one of the preliminaries to working on other amino acids. This is due to the facts (1) that acid-hydrolysed proteins, from which it is desired to remove certain amino acids chemically, have suffered a practically complete loss of tryptophane, and (2) certain proteins of interest because of natural deficiencies are also low in tryptophane content.

The chick diet used was a modification of a simplified chick diet which has been developed especially for work with proteins. The diet contained, per 100 g: casein, washed or acid hydrolysed, 20; gelatin, 10; calcium gluconate, 5; gum arabic, 5; tricalcium phosphate, 2; dipotassium phosphate, 0.5; potassium chloride, 0.5; manganese sulphate, 0.1; magnesium sulphate, 0.2; sodium silicate, 0.2; sodium chloride, 0.5; choline chloride, 0.3; ox bile salts, 0.5; wheat germ oil, 3; cod liver oil, 1; cotton pulp, 5 g. The vitamin B complex was added in the form of thiamin, 1 mg; riboflavin, 1 mg; pyridoxine, 1 mg; calcium pantothenate, 3 mg. A hexane extract of alfalfa equivalent to 2 gm was added as a source of vitamin K. A small quantity of a special mineral mixture was also added to provide iron, 1 mg; copper, 0.5 mg; zinc, 0.5 mg; cobalt, .02 mg; and iodine, 0.4 mg. An extract of brewers' yeast prepared by extraction with 50% aqueous methanol was concentrated, made slightly acid with acetic acid, and clarified by chilling and filtration. This was used at a level equivalent to 5%. Cerelese (glucose) was then added to complete the balance of 100 g. The principal features of this diet are the elimination of cereals and their by-products, and the reduction of protein sources to casein and gelatin.

An experiment was conducted to determine whether casein at a level of 20% would supply sufficient tryptophane for optimum growth of chicks on this diet. Chicks were first reared for 1 to 1½ weeks on a practical diet, then selected for uniform weight and condition, divided into groups of equivalent average weight, and given the experimental diets.

Pure l-tryptophane was added to the diet at various levels and the

TABLE I.
Growth-promoting Effects of l-tryptophane Additions to a Chick Diet Containing 20% Whole Casein.

l-tryptophane added, %	Avg gain per chick, g	Gain per day, %
0.0	43.3	6.7
0.16	46.0	7.0
0.26	44.2	6.8
0.36	46.0	7.0

Each group contained 5 chicks with an average starting weight of 50 g. The experiment lasted 9 days.

growth rate of the chicks determined at frequent intervals for 6 to 9 days. Results of the first experiment are given in Table I. It is evident from this experiment that additions of l-tryptophane did not increase the rate of growth of the chicks. Recent tables of amino acid composition of proteins¹ give a value of 2.2% for the tryptophane content of casein and 0.0% for that of gelatin. The tryptophane content of the diet, therefore, could not be appreciably more than 0.44%. This appeared to be sufficient for a good rate of growth. The rate of growth shown by chicks on the experimental diet was approximately equal to that which would be made by the same chicks on the practical rearing diet.

Other experiments were planned to investigate lower tryptophane levels. Casein was completely hydrolysed by means of strong sulphuric acid, the acid removed by barium hydroxide, and the hydrolysate concentrated under reduced pressure, decolorized with activated

TABLE II.
Growth-promoting Effect of l-tryptophane Additions to a Tryptophane-deficient Chick Diet Containing 20% Acid-hydrolysed Casein.

l-tryptophane added %	Exp. 1*		Exp. 2†		Exp. 3‡	
	Avg gain per chick, g	Gain per day, %	Avg gain per chick, g	Gain per day, %	Avg gain per chick, g	Gain per day, %
.0	— 6.4	—2.4	— 7.6	—2.2		
.2	+12.0	+3.7	+14.3	+3.3		
.3					+19.7	+3.3
.4			+19.6	+4.6	+26.1	+4.2
.5					+33.3	+5.1
.6			+25.3	+5.3	+33.9	+5.2

*Each group contained 4 chicks with an average starting weight of 48 g. The experiment lasted 6 days.

†Each group contained 5 chicks with an average starting weight of 47 g. The experiment lasted 8 days.

‡Each group contained 5 chicks with an average starting weight of 65 g. The experiment lasted 8 days.

¹ Calvery, H. O., *Chemistry of the Amino Acids and Proteins* (edited by C. L. A. Schmidt), Charles C. Thomas, publisher, Baltimore, Maryland, first ed., p. 217.

carbon, mixed with cotton pulp and dried in a current of warm air. Colorimetric tests for tryptophane (Hopkins-Cole, Rosenheim) were negative. The mixture was placed in the diet in lieu of the whole casein and cotton pulp. Results with this diet are given in Table II.

It will be seen from Table II that chicks given only the basal diet suffered a distinct loss in weight. This loss was clearly indicated within the first 24 hours of the experiments. Evidently, there was no appreciable tryptophane reserve. On the other hand, growth took place in the groups receiving as little as 0.2% l-tryptophane in the diet. The tendency of the latter groups to grow was also clearly indicated within 24 hours in most cases, although there was some evidence that chicks did not become fully accustomed to the new diet in this length of time. The magnitude of the differences in growth rates would indicate that the basal diet was practically free of tryptophane but that no other serious deficiency existed.

Maximum rate of growth was attained at 0.5 to 0.6% of added l-tryptophane. This rate was substantially less than that of chicks on the whole casein diets. Such differences in growth-promoting values of whole as compared to hydrolysed casein for the chick have been previously reported^{2, 3} even in the case of enzymatic hydrolysis,³ and are, therefore, to be expected.

Summary. The l-tryptophane requirement for optimum growth of the young chick is approximately 0.5% of the diet.

13379

Similarities and Possible Relationships Among Viruses of Psittacosis, Meningopneumonitis, and Lymphogranuloma Venereum.

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Findlay, Mackenzie and MacCallum¹ have described a developmental cycle for the virus of lymphogranuloma venereum in the brains of infected mice and have pointed out that the developmental

² Klose, A. A., Stokstad, E. L. R., and Almquist, H. J., *J. Biol. Chem.*, 1938, **123**, 691.

³ Stokstad, E. L. R., *Poultry Science*, 1940, **19**, 42.

¹ Findlay, G. M., Mackenzie, R. D., and MacCallum, F. O., *Nature*, 1938, **141**, 877; *Trans. Roy. Soc. Trop. Med. and Hyg.*, 1938, **32**, 183.