

cortical secretion as the controlling mechanism for the release of antibody from lymphocytes.

The anamnestic reaction is one manifestation of this control.

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Pyridoxine and Tryptophane Metabolism in Rice Moth Larvæ (*Corcyra cephalonica* St.).

P. S. SARMA. (Introduced by C. A. Elvehjem.)

From the Nutrition Research Laboratories, Indian Research Fund Association, Coonoor, S. India.

Lepkovsky and Nielsen¹ showed that the urine of rats which were kept on a pyridoxine-deficient diet contained a yellow pigment, which turned green at neutral pH on the addition of ferric salts like ferric ammonium sulphate. Later, Lepkovsky, Roboz, and Haagen-Smit² isolated the yellow pigment by chromatographic adsorption procedure and identified it as xanthurenic acid. It disappeared from the urine of rats when they were transferred from the deficient diet to one rich in pyridoxine or when pyridoxine was given orally. This compound was first studied by Musajo,³ who found it in the urine of rats and rabbits fed high protein diets.

It has been shown by the author⁴ that pyridoxine is one of the growth promoting factors for the rice moth larvæ (*Corcyra cephalonica* St. Lep.). It was therefore of interest to determine whether the rice moth larvæ excreted any yellow colored compounds in the feces when they were kept on a pyridoxine-deficient diet containing added amounts of tryptophane.

The pyridoxine-deficient diet consisted of salt-extracted wheat flour, sugar, and salt mixture together with the vitamins thiamine, riboflavin, nicotinic acid, and calcium pantothenate in the proportion of 10, 5, 50, and 15 γ per g of diet respectively. Rice moth larvæ which were feeding on whole wheat for a period

of 10-12 days after hatching were removed, cleaned, weighed, and placed on the diets given in Table I. The color of the excreta was noted after one week.

These results indicate that on a pyridoxine-deficient diet containing additional amounts of tryptophane, the larvæ excreted yellow-colored feces. Further, it was found that the yellow color disappeared from the feces of the larvæ when the latter were transferred to diets rich in pyridoxine. The same changes in the color of the larval excreta were observed even when bigger larvæ, *i.e.*, larvæ which had fed on whole wheat for 15-20 days after hatching, were placed on the diets mentioned in Table I.

The following experiments were carried out to determine whether the incorporation of tryptophane in the various deficient diets caused the yellow-colored excreta. Rice moth larvæ were taken from whole wheat, divided into 6 equal batches of 20 larvæ and placed on the diets given in Table II.

The results clearly show that only those larvæ which were placed on a diet deficient in pyridoxine excreted yellow-colored feces.

Seven amino acids, cystine, tryptophane, lysine, monohydrochloride, histidine, *l*-leucine, *d*-arginine, and tryosine were fed to the rice moth larvæ at a level of 50 mg of the amino acid to every 5 g of the pyridoxine-deficient diet. It was found that only tryptophane caused the excretion of the yellow-colored feces.

These results clearly indicate that there is an intimate relationship between pyridoxine and tryptophane metabolism in the rice moth larvæ.

The minimum amount of tryptophane

¹ Lepkovsky, S., and Nielsen, E., *J. Biol. Chem.*, 1942, **144**, 135.

² Lepkovsky, S., Roboz, E., and Haagen-Smit, A. J., *Ibid.*, 1943, **149**, 195.

³ Musajo, L., *Chem. Abst.*, 1935, **29**, 6292; *Boll. Soc. ital. biol. sper.*, 1935, **10**, 290.

⁴ Sarma, P. S., *Ind. J. Med. Res.*, 1943, **31**, 165.

TABLE I.

Diet	Color of the larval excreta	Wt in mg for 10 larvæ	
		Initial	After 2 weeks
1. Pyridoxine-deficient diet + 10 mg tryptophane per g diet	yellow	4.40	12.20
2. Pyridoxine-deficient diet + 5 mg tryptophane per g diet	yellow	4.35	13.24
3. Basal diet + all vitamins + 5 mg tryptophane per g diet	white	4.61	59.32
4. Basal diet + all vitamins	white	4.50	61.15
5. Pyridoxine-deficient diet	white	4.44	13.12

TABLE II.
Weight of Larvæ and Color of Excreta on Different Diets.

Diet	Color of the excreta	Wt in mg for 10 larvæ	
		Initial	After 2 weeks
1. 5 g riboflavin-deficient diet + 50 mg tryptophane	white	3.60	8.80
2. 5 g nicotinic acid-deficient diet + 50 mg tryptophane	"	3.70	37.23
3. 5 g basal diet + all vitamins + 50 mg tryptophane	"	3.50	38.91
4. 5 g pyridoxine-deficient diet + 50 mg tryptophane	yellow	3.50	8.22
5. 5 g pyridoxine-deficient diet + 100 mg tryptophane	"	3.72	9.56
6. Basal diet + all vitamins	white	3.75	36.63

which had to be added to the pyridoxine-deficient diet to produce a distinctly yellow-colored larval excreta was found to be about 3 mg per g of the diet.

The yellow pigment was found to be readily soluble in water, slightly soluble in aqueous acetone and aqueous alcohol, but insoluble in ether, absolute alcohol, benzene, chloroform, and acetone. The addition of mercuric sulphate solution (15%) caused its precipitation, as a mercury complex compound, but it was found difficult to purify the pigment and concentrate it, since hydrogen sulphide, which was used to remove the mercury, destroyed it.

However, the yellow pigment could be adsorbed on fuller's earth (B.D.H.) in acid medium and could also be easily eluted by treatment with dilute alkali solution. A concentrated solution was prepared in this manner, but attempts at obtaining the pigment in a pure crystalline form proved unsuccessful.

In aqueous solution the yellow pigment did not turn green on the addition of ferric ammo-

nium sulphate at neutral pH. Thus it does not show the most characteristic reaction of xanthurenic acid. Probably we are dealing with a pigment entirely different from xanthurenic acid and further investigation of its occurrence in larval excreta may throw more light on pyridoxine and tryptophane metabolism.

Summary. A yellow-colored compound is excreted by rice moth larvæ, when they were fed pyridoxine-deficient diets containing tryptophane. The yellow color disappeared from the excreta either on the inclusion of pyridoxine in the diet or on the transfer of the larvæ to a pyridoxine-rich diet. Of the 7 amino acids examined, only tryptophane gave rise to the yellow-colored excreta. The pigment is soluble in water, adsorbed on fuller's earth, but does not turn green with ferric ammonium sulphate at neutral pH. The compound in question may, therefore, be different from xanthurenic acid.