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# ABSORPTION OF THIAMINE AND NICOTINIC ACID IN THE RAT INTESTINE DURING FASTING AND IMMOBILIZATION STRESS

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Translation of "Vsmoktuvannya tiaminu ta nikotynovoy kysloty v kyshechnyku shchuriv pry holoduvanni ta immobilizatsiynomu stresi," Fiziolohichnyy Zhurnal, Vol. 23, No. 1, 1977, pp 98-102


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16. Abstract

By perfusion of isolated sections of intestine with a solution containing thiamine at a concentration of 3.1 µmole, it was established that thiamine absorption in animals fasted for 72 hours decreased by 28%, whereas absorption increased by 12% in rats after 24 hour immobilization. Besides, after immobilization, absorption of label in the intestinal mucosa increased. Na-K-ATPase activity in the intestinal mucosa increased. Na-K-ATPase activity in the intestinal mucosa decreased by 10% during fasting, and it increased with immobilization of the animals. Evidently, Na-K-ATPase activity in the intestinal mucosa cells determined the absorption rate of thiamine and nicotinic acid at the level of vitamin transport through the plasma membranes of the enterocytes.

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# ABSORPTION OF THIAMINE AND NICOTINIC ACID IN THE RAT INTESTINE DURING FASTING AND IMMOBILIZATION STRESS

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One possible cause of endogenous vitamin deficiency is disturbance /98* of the absorption of vitamins in the intestine [10, 20]. Clinically, the syndrome described is absorption deficiency, which is associated with many diseases [1, 9]. However, the characteristics of vitamin absorption in the intestine in pathologies, as well as the role of separate processes which provide vitamin transport through the intestinal cell membranes, have been studied insufficiently.

We studied the characteristics of absorption of thiamine and nicotinic acid in the small intestine of the rat during fasting and in animals under immobilization stress. At the same time, the $Na^+$-$K^+$ ATPase activity of the intestines of the experimental animals, as a membrane enzyme, was studied [6, 13]. This provides active transport of some substances through the membranes of the epithelial cells of the intestine [4, 12].

## Method

The studies were conducted on 50 white male rats, weighing 200±20 g, which were maintained on normal vivarium rations. The characteristics of thiamine and nicotinic acid absorbtion were studied under conditions of immobilization stress and fasting.

*Numbers in the margin indicate pagination in the foreign text.

The animals were immobilized for 24 hours in the horizontal position, to prevent numerous ulcers in the stomachs of the animals. Upon incision, the stomachs frequently are filled with blood, which is characteristic of immobilization stress [17].

Rats of the other group were not fed for 72 hours (unlimited water) before the experiment. After this time, the weight of the rats decreased by an average of 15%, but the weight of the intestine was unchanged.

The absorption of thiamine in the rat intestine was studied, by means of our modification of a method of perfusion of the intestine [14]. Under hexenal anesthesia, the intestinal wall of the rat was cut, and a section of the intestine 5 cm long was isolated, at the level of the upper third of the small intestine, without damaging the mesentery. The isolated section was perfused with a 0.9% sodium chloride solution, which contained thiamine at a concentration of 3.1 μM. The perfusion rate was 0.75 mℓ/min. The perfusate was collected fractionally every 2 min for a period of 10 min. Thiamine absorption was determined from the decrease in thiamine in the perfusate. The thiamine was determined in the tests according to [3].

Nicotinic acid absorption was determined from the degree of disappearance of the $^{14}C$ label, in administration of $^{14}C$ labelled nicotinic acid to a 5 cm long section of the small intestine, isolated at the level of the upper third of the small intestine. A 0.9% solution of sodium chloride which contained $^{14}C$ nicotinic acid at a concentration of 40 μM (activity of the preparation, 10 mC/g) was administered to the section in the amount of 1 mℓ. After 10 and 60 min, the solution was

separated from the lumen of the intestine, and the degree of decrease of the label in the contents of the section of intestine and the amount of label in the mucosa were determined. The radioactivity of the samples was determined in a PST-100 end counter. The results were expressed in count/g/min.

The $Na^+$-$K^+$ ATPase activity was determined by the method proposed by Govorova et al [2]. The results were expressed in μg phosphorus/g tissue/hour.

The results were processed biometrically [5].

Results

Study of the absorption of thiamine by a section of the small intestine of the rat, during its perfusion for 10 min with a thiamine solution (3.1 μM), permitted determination that, after 24 hour immobilization of the animals, absorption of thiamine from the perfusate increases by 12% (Fig. 1). While, in the control group of animals, of the total amount of thiamine (7.5 μg), 4.25 μg of thiamine was absorbed in 10 min, it was 5.25 μg in the rats which were immobilized. On the contrary, in the animals which were fasted for 72 hours, thiamine absorption decreased by 28% and, of the total amount of thiamine (7.5 μg), only 2.25 μg was absorbed (Fig. 1). /99

The change in the thiamine absorption rate, which was determined at 2 min intervals over 10 min, is in the same direction as that of the data presented (Fig. 2). In immobilization stress, the thiamine absorption rate increases to 0.55 μg/min, and it decreases to 0.23 μg/min in the animals which were fasted, with a standard 0.45 μg/min of thiamine in the perfused section of the small intestine. /100

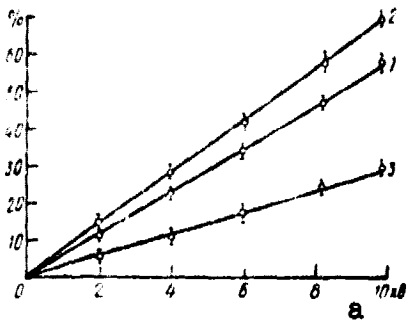

Fig. 1. Absorption of thiamine in rat intestine (in % of total amount in perfusate): 1. control animals; 2. animals under immobilization stress; 3. animals fasted for 72 hours.

Key: a. min

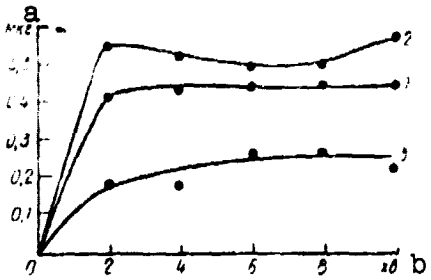

Fig. 2. Rate of absorption of thiamine in rat intestine (in μg thiamine per min): conventional designations as in Fig. 1.

Key: a. μg b. min

We determined nicotinic acid absorption in the small intestine of normal rats and in animals which endured immobilization stress. The study results are presented in Fig. 3. They characterize the degree of decrease of the $^{14}C$ label in the luminal contents, 10 and 60 min after administration of the $^{14}C$ nicotinic acid solution to the lumen of the intestine. A higher degree of absorption of nicotinic acid from the lumen of the intestine of the rat during immobilization stress was found. The difference in the decrease in label was a little higher in 10 than in 60 min.

Fig. 4 presents the results of study of the accumulation of the $^{14}C$ label of nicotinic acid in the mucosa of the section of intestine of normal rats and those which endured immobilization stress. A sharp increase in accumulation of label in 10 min was traced in the intestinal mucosa of the rats which endured immobilization. Besides, the difference in accumulation of label in the mucosa was less pronounced in 60 min, although the direction of the process is the same.

Study of $Na^+$-$K^+$ ATPase showed that the activity of the enzyme in the intestinal mucosa of the rat depends on the condition of the body

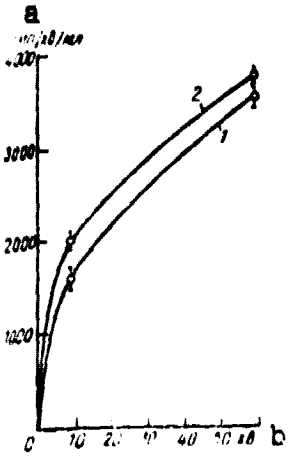


Fig. 3. Nicotinic acid absorption from rat intestinal lumen: conventional designations as in Fig. 1.

Key: a. count/min/mℓ
b. min

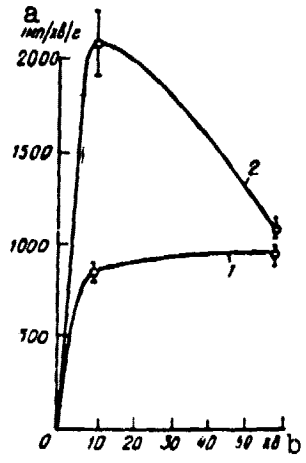


Fig. 4. Nicotinic acid accumulation in rat intestinal mucosa upon absorption from intestinal lumen: conventional designations as in Fig. 1.

Key: a. count/min/g
b. min

of the animal. Fig. 5 presents the results of determination of $Na^+$-$K^+$ ATPase activity in the intestinal mucosa of the rat under immobilization stress and in fasted animals, compared with normal rats. In 24 hour immobilization, the activity of the enzyme under study increased sharply, and it is nearly 50% above the $Na^+$-$K^+$ ATPase activity of the mucosa of the small intestine of normal animals. In 72 hour fasting, a tendency develops toward a decrease in activity of the enzyme, compared with the group of control animals.

## Discussion

It was determined from the study that, under immobilization stress conditions, thiamine and nicotinic acid absorption in the rat intestine increases. At the same time, $Na^+$-$K^+$ ATPase activity in the mucosa of the upper third of the small intestine of the rat increases.

According to the data of Tursunkhodzhayeva et al [8], the consumption of group B vitamins increases under stress. It follows from the data we obtained that the increase of vitamin consumption is produced by increase

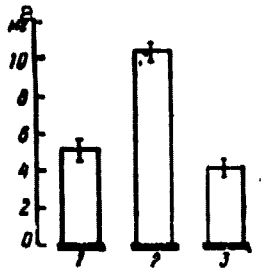


Fig. 5. $Na^+$-$K^+$ ATPase activity in mucosa of rat small intestine: conventional designations as in Fig. 1.

Key: a. mg

in absorption in the intestine, due to an increase in activity of $Na^+$-$K^+$ ATPase, which participates in transmembrane transport of matter [4].

It is known that $Na^+$-$K^+$ ATPase is a membrane fragment [6], localized chiefly in the lateral and basal plasma membranes of the enterocytes [7, 13]. The entry of thiamine into the blood and lymph apparently is connected with the functioning of this enzyme [18]. It is known that the activity of this enzyme is under hormonal control, since the possibility of activation of $Na^+$-$K^+$ ATPase in some tissues of animals by corticosteroids [13], the content of which increases under stress [11], has been established.

The decrease in thiamine absorption in fasting animals must be explained by decreases in $Na^+$-$K^+$ ATPase activity in the intestinal mucosa. It must be noted that some authors point out a decrease in activity of the intestinal enzymes which participate in digestion and absorption, when peroral intake of foodstuffs to the body is stopped [15], or in parenteral feeding [16], which is accompanied by a decrease /101 in the absorptive capacity of the intestine.

Therefore, the absorption of thiamine and nicotinic acid in the rat intestine in small physiological concentrations (3.1 and 40 μM) depends on the activity of membrane $Na^+$-$K^+$ ATPase, which provides active transport through the plasma membrane of the cells of the intestine [12]. A reduction in activity of this enzyme by hormones or other actions (anorexia, exclusion of the functional activity of the intestine)

affects the level of absorption of the coenzyme vitamins thiamine and nicotinic acid in the intestine which, in the final analysis, can affect the provision of the body with these vitamins.

## REFERENCES


1. Vinogradova, M.A., "Absorption deficiency syndrome in some internal diseases," author's abstract of dissertation, Moscow, 1968.

2. Govorova, L.V., A.Ye. Aleksandrova and S.E. Teplov, "Change of ATPase activity of the brain and erythrocytes in anoxia," Voprosy med. khimii 21, 23-26 (1975).

3. Yeliseyeva, G.D., "Fluorometric determination of thiamine, cocarboxylase and riboflavin in biological objects," in the collection Vitaminy [Vitamins], Kiev, 1953, 1, pp. 38-57.

4. Lisovskaya, N.P., "Cell membrane adenosine triphosphatase and ion transport," in the collection Uspekhi biol. khimii [Progress in Biochemistry], Moscow, 1967, 8, pp. 93-116.

5. Plokhinskiy, N.A., Biometriya [Biometrics], Moscow, 1970.

6. Rid, E., "Membrane enzymes," in the book Tsitologiya fermentov [Cytology of Enzymes], Moscow, 1971.

7. Tashmukhamedov, B., A. Dalimova and Z. Bekmukhamedova, "Study of the localization of transport ATPase in the small intestine," Biofizika 14, 1032-1034 (1969).

8. Tursunkhodzhayeva, M.S. and A. Shadmanov (cited in [10]).

9. Faytel'berg, R.O., Vsasyvaniye uglevodov, belkov i zhirov v kishechnike [Absorption of Carbohydrates, Proteins and Fats in the Intestine], Leningrad, 1967.

10. Khmelevskiy, Yu.V. and A.Ya. Rozanov, Obmen vitaminov pri serdechno-sosudistyky zabolevaniy [Vitamin Metabolism in Cardiovascular Diseases], Zdorov'ya Press, Kiev, 1975.

11. Eskin, I.A., Osnovy fiziologii endokrinnykh zhelez [Foundations of the Physiology of the Endocrine Glands], Moscow, 1975.

12. Csaky, T.Z., "A possible link between active transport of electrolytes and nonelectrolytes," Fed. Proc. 22/1, 3-7 (1963).

13. Fujita, M., H. Matsui, P. Nagano and M. Nakao, "Asymmetric distribution of ouabain-sensitive ATPase activity in rat intestinal mucosa," Biochim. et Biophys. Acta 233, 404-408 (1971).

14. Hara, J., H. Wakasugi and I. Takahashi, "Study on intestinal absorption of sugar in thiamine and riboflavin deficiency," Vitamins 37, 284-288 (1968).

15. Heitanen, E., "The effect of different diets on the alkaline phosphatase, ATPase and disaccharidase levels of the small intestine of the rat," Comp. Biochem. Physiol. 150, 41-46 (1975).

16. Levine, G., L. Deren, E. Steiger and R. Zinno, "Role of oral intake in maintenance of gut muss and disaccharidase activity," Gastroenterology 67, 975-982 (1974).

17. Menguy, R. and J. Masters, "Gastric mucosal energy metabolism and 'stress ulceration'," Ann. Surg. 180, 538-546 (1974).

18. Rindi, G. and U. Ventura, "Thiamine intestinal transport," Physiol. Rev. 52, 821-827 (1972).

19. Srastny, F., "Corticosteroid regulation of the $Na^+$-$K^+$ ATPase activity in the chick embryo cerebral hemispheres," Ontogenesis Brain, Prague, 1974, pp. 87-98.

20. Thomson, A., H. Baker and C. Leevy, "Patterns of $S^{35}$ thiamine hydrochloride absorption in the malnourished alcoholic patients," J. Lab. Clin. Med. 76, 34-45 (1970).