of histologic preparations showed that the morphologic pattern of the alimentary canal as a whole corresponded to that described above for intact animals, and the epithelial cells were not damaged. Changes observed were evidence only of activation of digestive processes. To confirm this observation, we used a luminescence-serologic method with antiserum to NAG vibrios. The investigation showed that antigenic material of the bacteria in each case and the bacterial cells themselves were revealed by their luminescence in the lumen of the alimentary canal of the daphnias 3 h after the experiment began. Intense fluorescence of the peritrophic membrane of the middle portion of the alimentary canal also was observed, where the introduced microorganisms were being digested (Fig. 3a). Fluorescence was found mainly in the peritrophic membrane of the posterior portion 48 h after introduction of the bacteria into the habitat of the crustaceans, whereas in the middle portion fluorescence of the striated border and cytoplasm of the epithelial cells also was observed (Fig. 3b). On the 3rd day of the experiment specific fluorescence could no longer be observed in any portion of the digestive tract of the daphnias.

The methods of investigation which we used showed that introduction of enteropathogenic microorganisms into the habitat of these crustaceans gives rise to no pathological changes in the alimentary tract of daphnias. The absence of phenomena of phagocytosis in daphnias, observed and described by Mechnikov [3] in his classical research, in our experiments can be explained on the grounds that their intestinal epithelium is sufficiently well protected from mechanical injury by a peritrophic membrane, a peritrophic state, where food is digested, and a chitin membrane. The method of peroral infection which we used did not enable the leukocytes of the daphnias to come into contact with the pathogenic vibrios. In addition the epithelial cells of the alimentary canal of crustaceans evidently do not possess receptors for toxins produced by bacteria of the enteric group, and these animals accordingly do not react to them.

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EFFECT OF α-TOCOPHEROL (VITAMIN E ACETATE) ON METABOLISM OF GASTRIC SECRETORY CELLS OF HYPERTHYROID RATS

S. N. Fedchenko

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The leading factor in the complex chain of adaptive reactions of the stomach to an excessive intake of thyroid hormones is modification of the permeability of the biological membranes [3, 4, 10]. It has been shown that thyroid hormones activate lipid peroxidation (LPO) in the liver mitochondria [2]. The writer previously [5, 6] described structural disturbances in mitochondria and lysosomes in the gastric parietal cells of hyperthyroid rats. At this stage it is very important to look for possible ways of influencing metabolism in

Department of Biology, Voroshilovgrad Medical Institute.

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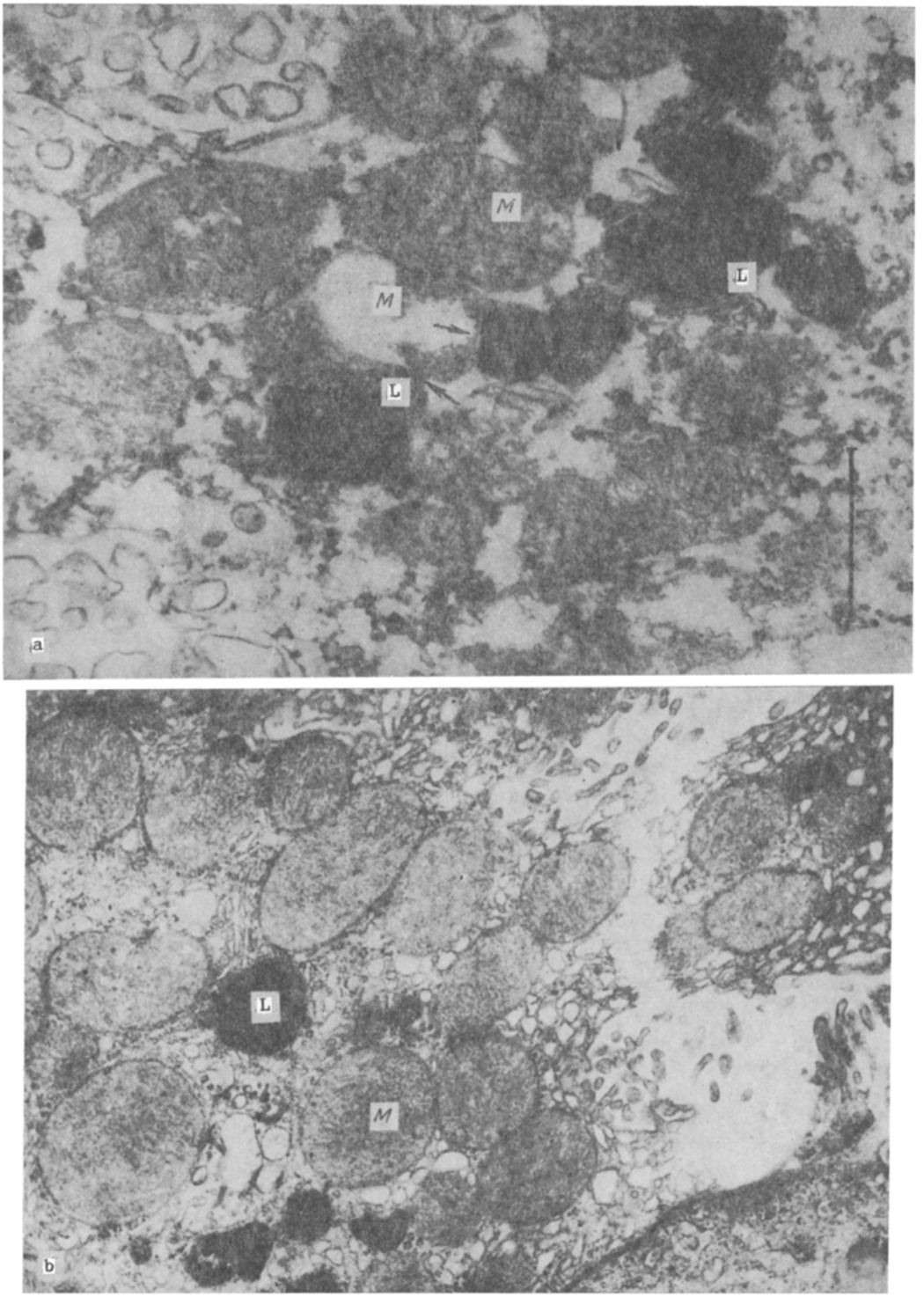

Fig. 1. Fragment of gastric parietal cell of hyperthyroid rat not receiving TP (a) and of hyperthyroid rat receiving injection of TP (b). a) Increase in number of lysosomes, mitochondria (M) structurally incomplete; electron-histochemical reaction product for AP localized in lysosomes (L). Arrow indicates outflow of enzyme from lysosomes. 40,000 ×; b) stabilization of mitochondrial (M) and lysosomal (L) membranes. 32,000 ×.

in the gastric mucosa in order to increase the resistance of the membranes. An interesting substance in this connection is α-tocopherol (TP).

Accordingly, in the investigation described below, the effect of TP was studied on mitochondrial and lysosomal membranes in gastric secretory cells in animals saturated with thyroxine.

TABLE 1. Effect of TP on Enzyme Activity (in relative units) in Gastric Parietal Cells of Hyperthyroid Rats (M ± m)

Group of animals	SDH	NDH	CCO	G6PDH	LDH
Control	70,3±1,68	66,7±3,4	20,1±0,11	43,8±1,0	42,5±1,18
1	88,8±2,37*	89,0±2,7*	27,3±0,14*	64,9±2,16*	49,6±0,98
2	78,1±1,8	81,0±2,6	24,4±0,11	51,7±2,11	40,4±1,14
3	44,2±2,0*	41,3±0,4*	11,4±0,08*	76,1±1,20*	68,7±1,32*
4	64,1±2,13	54,4±0,31	17,1±0,07	45,1±2,12	40,8±1,64

Legend. Here and in Table 2: *P < 0.05 compared with control.

TABLE 2. Results (in relative units) of Morphometric Analysis of Parietal Cells in Gastric Mucosa of Rats (M ± m)

Group of animals	Parameter		
	S_{mc}	S_{cr}	K
Control	32,5±0,43	2,9±0,029	0,57±0,027
1	39,6±0,51*	3,5±0,018*	0,55±0,011
2	34,3±0,49	3,1±0,011	0,50±0,008
3	40,1±0,41*	2,0±0,014*	0,98±0,07*
4	36,4±0,39*	2,5±0,017	0,69±0,013

Legend. S_{mc}) fraction of area of section through cell cytoplasm occupied by mitochondria; S_{cr}) surface area of mitochondrial cristae as a fraction per unit volume of cytoplasm; K) coefficient of fragmentation of cristae.

EXPERIMENTAL METHOD

Experiments were carried out on 78 male Wistar rats weighing 180-200 g. Sixteen rats, kept under animal house conditions, served as the control. The remaining animals were divided into four groups: 14 rats of group 1 were given L-thyroxine intraperitoneally in a dose of 2.5 mg/kg daily for 10 days, 14 rats of group 2 received, besides thyroxine by the scheme described above, TP (15% oily solution) in a dose of 1 mg/kg daily for 10 days, 16 rats of group 3 received thyroxine in a dose of 2.5 mg/kg for 30 days, and the 18 rats of group 4, besides thyroxine by the scheme described above, received TP for 30 days. Activity of succinate dehydrogenase (SDH), cytochrome oxidase (CCO) by the method in [8], NADH-dehydrogenase (NDH), glucose-6-phosphate dehydrogenase (G6PDH), and lactate dehydrogenase (LDH), by the method in [9], was determined histochemically and quantified cytophotometrically. The surface area of the mitochondrial cristae, the coefficient of their fragmentation, and the degree of swelling of the mitochondria were calculated and the number of lysosomes estimated by the method in [1]. Acid phosphatase (AP) was detected electron-cytochemically by the method in [7].

EXPERIMENTAL RESULTS

Administration of thyroxine was accompanied by marked changes in metabolism in the gastric secretory cells and, in particular, in the parietal cells. This was shown by increased SDH, NDH, and CCO activity on the 10th day of the experiment by 26, 32, and 35% respectively (Table 1). The surface area of the mitochondrial cristae also was increased (Table 2).

A sharp increase in activity of these dehydrogenases could be connected with swelling of the mitochondria or increased membrane permeability, evidence of an increase in the functional load on the parietal cells and, in our opinion, a compensatory response to excess administration of thyroxine, with activation of the Krebs cycle and the terminal electron transport chain, one of the most energy-consuming pathways of glucose oxidation.

Electron-microscopic analysis of the parietal cells showed an increase in the number of swollen mitochondria, hyperosmosis of their matrix, an increase in the number of cristae and in their surface area to 3.5 relative units compared with 2.9 in the control, but no change in the coefficient of fragmentation of the mitochondrial cristae (Fig. 1, Table 2).

After administration of thyroxine for 30 days SDH, NDH, and CCO activity was reduced in the parietal cells, evidence of exhaustion of their enzyme systems. As a compensatory reaction to maintain the energy balance of the cell the intensity of anaerobic glycolysis increased and LDH activity rose by 64% and G6PDH activity by 72% of their initial level (Table 1). Since intensification of glycolysis under conditions of long-term hyperthyroidism is accompanied by accumulation of incompletely oxidized products, this guarantees glycolysis as a source of energy.

Ultrastructural analysis of the gastric parietal cells of rats receiving thyroxine for 30 days showed that in many of them structurally incomplete mitochondria were formed, with a reduced number of cristae; the surface area of the mitochondrial cristae was reduced by 1.4 times and the coefficient of fragmentation of the cristae increased by 1.7 times (Table 2). At these same times an increase in the number of myelin figures and lysosomes and destruction of their membranes, with release of the enzyme AP, were observed (Fig. 1).

Administration of TP together with thyroxine was accompanied by less marked structural changes in the mitochondria and lysosomes (Table 2).

Activity of the enzymes of glycolysis remained within the control limits (Table 1). Stabilization of mitochondrial (a decrease in the coefficient of fragmentation of the cristae) and lysosomal membranes was observed. This can be interpreted as the result of correction of the increased rats of LPO by TP, and in turn, this helped to reduce permeability of the mitochondrial and lysosomal membranes.

The results show that milder structural and metabolic changes develop under the influence of TP in the gastric parietal cells of hyperthyroid rats, evidence of the beneficial effect of this compound on membrane permeability.

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