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EFFECT OF SYNTHETIC PHENOLIC ANTIOXIDANTS ON THE JUXTAMURAL pH OF THE GASTROINTESTINAL TRACT OF INTACT AND VAGOTOMIZED RATS

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The aim of this investigation was to study the effect of synthetic inhibitors of free-radical oxidation reactions belonging to the group of sterically screened phenols, fat-soluble (dibunol — 4-methyl-2,6-di-tert-butylphenol) and water-soluble [2,6-di-tert-butyl-4-(1-aminoethyl)phenol hydrochloride — BH-3], on the juxtamural pH (JpH) profile of the digestive tract in rats under normal and pathological conditions. Such an investigation is necessary because in recent years antioxidants (AO) have begun to be used for the treatment of diseases of the digestive system [1, 5-7], whereas their effect on the structure and functions of the digestive organs under normal and pathological conditions has not been studied. As the criterion for evaluation of the action of AO on function of the gastrointestinal tract, we used a physiological parameter, namely the hydrogen ion concentration in the paramucosal (juxtamural) layer (JpH). The pathological state of the digestive system was simulated by subdiaphragmatic division of the vagus nerves. When this model was chosen the following factors were taken into consideration: 1) the craniobulbar portion of the CNS, represented by the vagus nerves, is the main source of parasympathetic innervation of the digestive organs [9]; 2) vagotomy is widely used in the surgical treatment of duodenal ulcer [12].

EXPERIMENTAL METHOD

Experiments were carried out on 150 male albino rats weighing 150-210 g. In series I the experiments were performed on initially intact animals (n = 103), of which 21 rats received a single injection of high doses of the compounds (45 mg/kg dibunol in a 3% solution of Tween-80 and 40 mg/kg of BH-3 in physiological saline, intraperitoneally), and the investigation was carried out 24 h later; 68 rats received the compounds in therapeutic doses (20 mg/kg dibunol and 25 mg/kg BH-3) and were investigated 7, 15, and 30 days after the first injection (the doses of the compounds were recommended by the Sector of Kinetics of Chemical and Biological Processes, Institute of Chemical Physics, Academy of Sciences of the USSR); in the control, 14 rats received 1 ml of a 3% solution of Tween-80 intraperitoneally, daily for 8 days. In series II rats subjected to bilateral subdiaphragmatic truncal vagotomy served as the test object. Starting from the 2nd day after the operation they were given the compounds in therapeutic doses and were investigated 7, 15, and 30 days after vagotomy. Under urethane anesthesia (150 mg/100g) JpH of the experimental and control animals of both series was measured 24 h after the last injection of the compound and after feeding, in the fundus of the stomach, the duodenum, jejunum, ileum, cecum, and rectum, by the method in [8]. Values of the gastroduodenal and ileocecal gradients (the coefficients, E_1 and E_2) were

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TABLE 1. Effect of Dibunol and BH-3 on JpH of the Alimentary Canal of Intact (Int) and Vagotomized (Vag) Rats (M ± m)

Experimental conditions	Time of investigation, days	Stomach	Duodenum	Jejunum	Ileum	Cecum	Rectum
Int		5,78±0,06	7,92±0,04	8,11±0,08	8,89±0,06	10,05±0,08	8,57±0,03
Int + Tween-80		5,74±0,09	7,76±0,31	8,06±0,11	8,84±0,11	9,88±0,23	8,68±0,12
Int + dibunol	1	5,83±0,22	7,80±0,12	8,07±0,09	8,86±0,09	9,84±0,19	8,53±0,22
Int + BH-3		6,65±0,13**	7,86±0,21	8,03±0,23	8,87±0,11	10,2±0,21	8,76±0,09
Int + dibunol	7	5,48±0,28	7,99±0,09	8,20±0,05	8,90±0,09	10,15±0,1	8,49±0,06
Int + BH-3		6,21±0,31	7,73±0,12	7,99±0,05	8,85±0,08	9,90±0,16	8,95±0,13
Vag		7,85±0,14*	7,63±0,08*	7,94±0,06	8,44±0,09*	9,36±0,11*	8,32±0,11
Vag + dibunol		7,57±0,45	7,62±0,12	7,94±0,11	8,45±0,14	9,35±0,27	8,38±0,21
Vag + BH-3		7,06±0,38	7,10±0,08	7,65±0,09	8,46±0,12	9,38±0,13	8,47±0,24
Int + dibunol	15	6,17±0,21	8,12±0,12	8,18±0,06	9,05±0,11	9,60±0,16	8,35±0,08
Int + BH-3		5,79±0,19	8,05±0,11	8,19±0,2	9,09±0,07	9,89±0,24	8,66±0,18
Vag		7,98±0,83	7,89±0,19	8,16±0,11	8,41±0,09*	9,43±0,22	7,70±0,45
Vag + dibunol		7,97±0,21	8,25±0,18	8,45±0,18	8,44±0,38	10,07±0,49	8,03±0,56
Vag + BH-3		8,72±0,59	7,90±0,38	8,59±0,25	8,48±0,31	10,19±0,49	8,24±0,41
Int + dibunol	30	5,78±0,26	8,02±0,18	7,91±0,15	8,74±0,13	9,88±0,19	8,63±0,09
Int + BH-3		5,71±0,18	7,75±0,16	7,86±0,15	8,43±0,2	9,78±0,23	8,30±0,19
Vag		4,98±0,33	7,66±0,04	7,78±0,07	8,34±0,08	9,30±0,09*	8,33±0,08
Vag + dibunol		5,97±0,41	7,55±0,07	7,80±0,08	8,36±0,13	9,29±0,27	8,74±0,30
Vag + BH-3		6,04±0,29	7,42±0,18	8,03±0,06	8,58±0,07	9,39±0,15	8,49±0,09

Legend. *$p < 0.05$ compared with intact rats, **$p < 0.05$ compared with vagotomized rats.

calculated as the ratios between the values of JpH in the corresponding segments of the digestive tube. The results of the measurements were subjected to statistical analysis [10].

EXPERIMENTAL RESULTS

The results show that a proximal-distal gradient of JpH exists in the alimentary canal of intact rats, and is characterized by the presence of two local JpH drops: gastroduodenal from acid to weakly alkaline ($E_1 = 0.70 \pm 0.005$) and ileocecal — from weakly alkaline to alkaline ($E_2 = 0.91 \pm 0.004$). A single injection of BH-3 in a high dose after 24 h caused JpH in the stomach to rise and this was accompanied by a decrease of the gastroduodenal JpH gradient ($E_1 = 0.81 \pm 0.01$, Table 1). The use of dibunol under the same conditions had no such effect. Chronic administration of the compounds was not accompanied at any of the times of testing by any significant change in the JpH profile of the gastrointestinal tract. Injection of the solubilizer Tween-80 likewise was ineffective. Besides the action of BH-3 on acid production in the stomach, which has been described, it also has an effect on the motor function of this organ. This takes the form of delayed evacuation of chyme from the stomach and the development of gastric stasis. This phenomenon was most marked after 15 days. At this time dibunol likewise had a similar action on the motor function of the stomach, but to a much lesser degree. Vagotomy led after 7 days to an increase in JpH in the stomach and a decrease in the duodenum, thus resulting in equalization of the gastroduodenal gradient ($E_1 = 1.004 \pm 0.02$), and also to a decrease in JpH in the ileum and cecum, with preservation of the physiological gradient. After 15 days JpH was observed to fall in the ileum, and after 30 days it increased in the stomach and decreased in the cecum. Injection of the compound into vagotomized animals caused no significant transformation of the JpH profile of the gastrointenstinal tract at any time of the investigation. As regards their effect on gastric motor function under these conditions, this was exhibited to a much lesser degree (BH-3 induced moderate gastric stasis after 15 days).

It can be concluded from generalization of these results that AO of the phenolic series (dibunol and BH-3), in therapeutic doses, have no significant effect on the JpH profile of the alimentary canal of intact rats and do not enhance changes in it caused by vagotomy. A detailed analysis of the post-vagotomy modification of the JpH relief of the gastrointestinal tract and of its possible mechanisms was given in [11]. Meanwhile, the present writers showed previously that vitamin E, in the form of α-tocopheryl acetate, induces a marked decrease in JpH in the stomach of both intact and vagotomized rats 7 days after injection [3]. At first glance, this fact contradicts the results of the present investigation. In

fact, if a natural AO as powerful as vitamin E stimulates secretion of H^+ by the glandular epithelium of the stomach, it may be concluded that the system of lipid peroxidation (LPO) is directly involved in this process. Meanwhile dibunol and BH-3, which surpass it in antioxidative activity, had no effect on acid production in the stomach. The reason for the different effects of α-tocopherol (TP) and the AO which we investigated on JpH in the stomach, in our view, is that vitamin E is a polyfunctional biologically active substance [4, 13, 14] and its effect on the gastric glands may be unconnected with its antioxidative properties. A similar phenomenon is found if the action of TP and dibunol on the rate of RNA biosynthesis in the liver is compared: the former stimulated it, and as has been shown, not by an antioxidative mechanism, whereas the second had no effect [2].

It can be concluded from this investigation that synthetic AO of the phenolic series, dibunol and BH-3, have no significant effect on the H^+ concentration in the juxtamural layer of the gastrointestinal tract of intact or vagotomized rats. This state of affairs indicates that these compounds can be subjected to experimental (and later, perhaps, clinical) trials as preparations for the pharmacotherapy of diseases of the digestive system in whose pathogenesis an essential role is played by nonphysiological activation of LPO. It must be recalled, under these circumstances, that BH-3 inhibits gastric motor function and may lead to the development of gastric stasis.

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