Effects of Taurine, Carnosine, and Casomorphine on Functional Activity of Rat Peritoneal Mast Cells

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We studied the effects of taurine, carnosine, and casomorphine on histamine release from rat peritoneal mast cells induced by compound 48/80 and ionophore A23187. Differences were revealed in the effect of the test preparations. Taurine inhibited histamine release induced by ionophore A23187, but not by compound 48/80. Carnosine abolished the stimulatory effect of compound 48/80 on histamine release, but did not modulate the effect of ionophore A23187. Casomorphine inhibited histamine release induced by ionophore A23187, but potentiated the effect of compound 48/80. The mechanisms for these effects are discussed.

Key Words: *mast cells; histamine; taurine; carnosine; casomorphine*

Published data show that mast cells (MC) play an important role in the anaphylactic reaction, inflammation, malignant growth, infectious processes, and allergic response [3]. MC not only synthesize and accumulate bioactive substances (heparin, histamine, *etc*.), but also release them by exocytosis during the immune response. Ca^{2+} ions play a key role in this process [7]. Secretion of substances from MC also accompanies the influence of nontoxic agents. Degranulation of MC induced by compound 48/80 or calcium ionophore A23187 is a convenient experimental model for studies of processes in membranes under conditions not associated with the cytotoxic effects of the test factor. Degranulation of MC is activated by endogenous bioactive compounds, including biogenic amines and peptides [2,8]. Histamine released during activation of MC is involved in the allergic response, particularly during treatment with various drugs. Study of MC exocytosis under the effect of compounds whose level undergoes considerable variations under the influence of food products or drugs is important for targeted correction of organism's sensitivity. Here we studied the *in vitro* effect of endogenous bioactive substances taurine, carnosine, and casomorphine on histamine secretion from rat peritoneal MC induced by compound 48/80 and ionophore A23187.

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MATERIALS AND METHODS

Function of MC was studied using purified fraction of peritoneal MC from male Wistar rats. The animals were decapitated under ether anesthesia. MC were isolated from the peritoneal fluid [15]. The purity of mast cell fraction reached 95%. Immediately after isolation the cells were resuspended in the medium, treated with taurine (2-aminoethanesulfonic acid, ICN Biomedical Inc.), carnosine (β-alanyl-L-histidine, Prof. S. L. Stvolinskii), or casomorphine (β-casomorphine-7, Institute of Molecular Genetics) in a concentration of 10^{-5} M, and incubated at 37°C for 10 min. Compound 48/80 (0.25 μg/ml) or calcium ionophore A23187 (0.125 μg/ml) was added to the incubation medium. The mixture was incubated for 5 min. Quantitative analysis of histamine was performed by the fluorometric method on a Hitachi-850 spectrofluorometer

[13]. Secretion of histamine in the absence (spontaneous secretion) or presence of taurine, carnosine, or casomorphine did not exceed 3-5%. These values were subtracted from the corresponding parameters in test samples. Antioxidant activity of compounds was estimated using the model system of quercetin autooxidation in the presence of N,N, N',N'-tetramethylethylenediamine at pH 10. $O_2^{\bullet}$ is an intermediate product of autooxidation [4]. The amount of oxidized quercetin was determined by optical density at 406 nm on a Hitachi-56 spectrophotometer. The effectiveness of the test substances was estimated by inhibition of quercetin autooxidation (%).The difference between optical densities in control and treated samples was divided by optical density in the control (λ=406 nm).

The results were analyzed statistically. The difference between control and treated samples was evaluated by Student's *t* test. The differences were significant at $p<0.05$.

RESULTS

Preincubation of MC with taurine (10^{-5} M) significantly inhibited A23187-induced histamine release (by 27%), but had no effect on cells treated with compound 48/80 (Fig. 1, *a*). Degranulation of MC under the influence of compound 48/80 is related to the release of intracellular Ca^{2+}. Taurine did not modulate MC degranulation induced by compound 48/80. The amount of intracellular taurine decreases, because the incubation medium does not include several factors necessary for active transport of taurine through the cell membrane (*e.g.*, pyridoxine). The concentration of taurine is high in marine mussels. Taurine is a sulfur-containing amino acid that plays an important role in the organism. Taurine is involved in the synthesis of bile acids, osmoregulation, and stabilization of membranes. Taurine has a positive effect during therapy of cardiomyopathy and other diseases, which is related to its antioxidant activity [8,14]. It cannot be excluded that taurine modulates function of ion channels and chelates bivalent ions [12]. The observed inhibition of A23187-induced degranulation requiring extracellular calcium in the presence of taurine can be explained by binding Ca^{2+} ions with taurine in the incubation medium, which results in a decrease in Ca^{2+}-dependent release of histamine from MC.

In contrast to taurine, dipeptide carnosine suppressed compound 48/80-induced histamine release from MC by 33%. However, no changes were revealed in cells treated with ionophore A23187 (Fig. 1, *b*). It was hypothesized that the therapeutic and preventive effects of carnosine in brain and cardiovascular pathologies are associated with its antioxidant activity [1]. Carnosine inhibits compound 48/80-induced activation of MC, which is probably related to detoxification of reactive oxygen species formed under the influence of compound 48/80

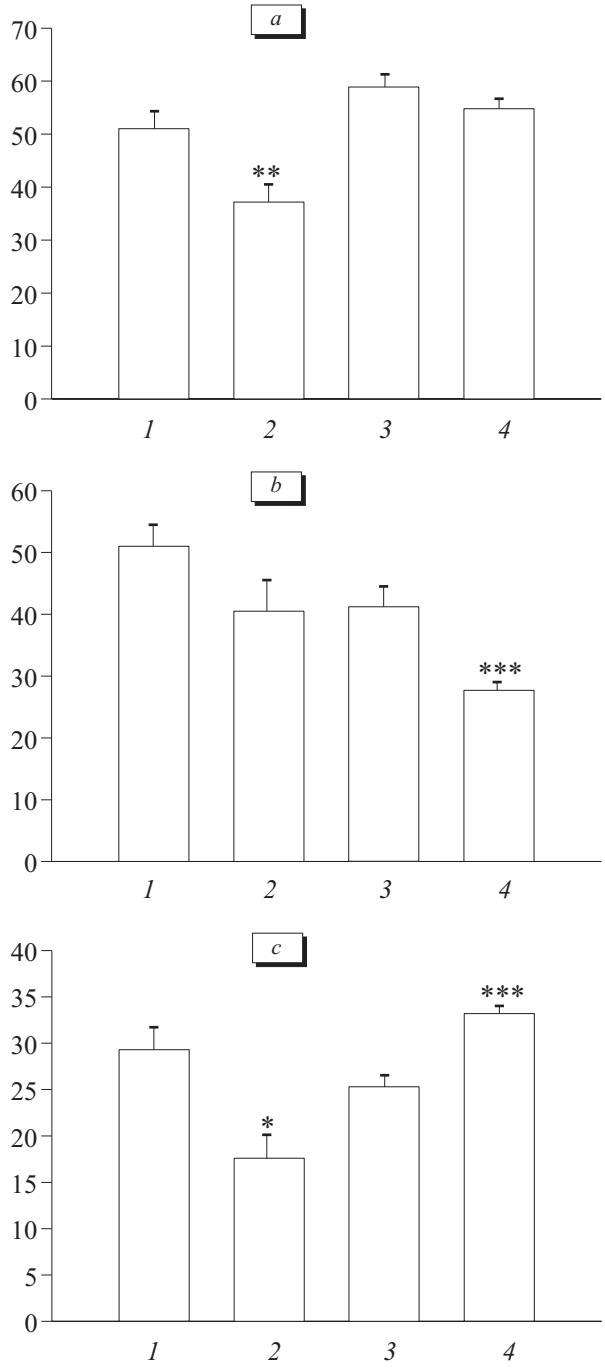

Fig. 1. Effects of taurine (*a*), carnosine (*b*), and casomorphine (*c*) on histamine release from rat peritoneal mast cells induced by ionophore A23187 and compound 48/80 (%). A23187 (*1*); taurine+A23187 (*2*); 48/80 (*3*); and taurine+48/80 (*4*). *$p<0.05$, **$p<0.02$, and ***$p<0.01$ compared to histamine release from the corresponding liberator (*1*, *3*) in the absence of preparations.

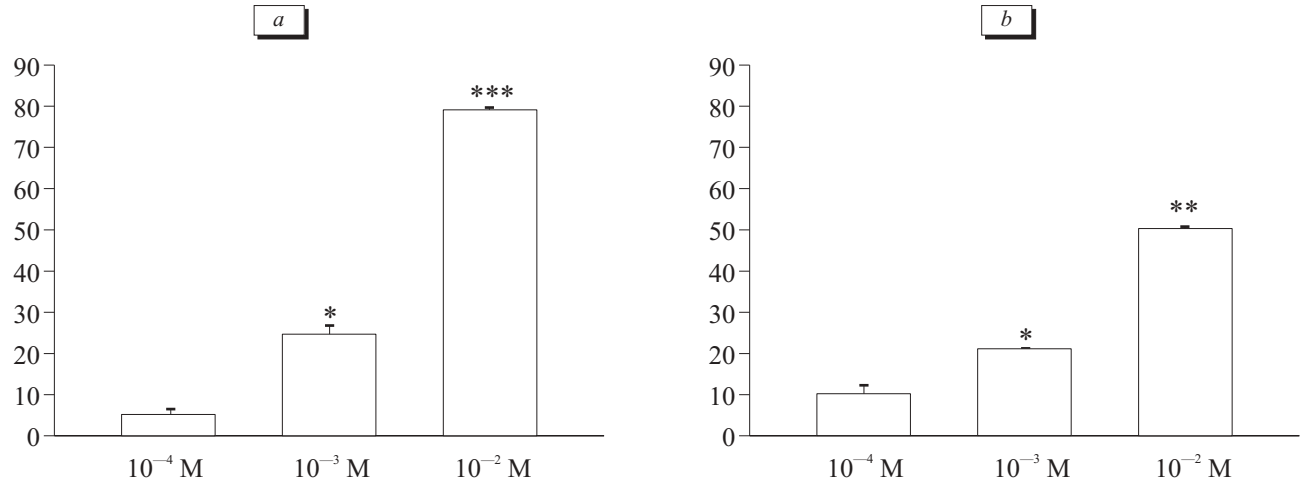

Fig. 2. Inhibition of quercetin autooxidation by taurine (*a*) and carnosine in various concentrations (*b*). Ordinate: % inhibition of quercetin autooxidation. **p*<0.05, ***p*<0.01, and ****p*<0.001 compared to test preparation in a concentration of 10^{-4} M.

[11]. Antioxidant activity of carnosine and taurine in doses of 10^{-4}-10^{-2} M was studied in the model system of quercetin autooxidation [4]. $O_2^{\bullet}$ serves as an intermediate product of autooxidation under these conditions. The test substances in concentrations of 10^{-3}-10^{-2} M exhibited strong inhibitory properties. Antioxidant activity of taurine surpassed that of carnosine (Fig. 2, *a*, *b*). It should be emphasized that the concentration of taurine in experiments with MC was 2 orders of magnitude lower. These data suggest that the protective effect of carnosine is related to interaction with cell receptors, but not to its antioxidant activity.

Polypeptide casomorphine is characterized by a wide range of biological properties [5,10]. Casomorphine inhibited ionophore A23187-induced histamine release from MC by 33%. This polypeptide potentiated the effect of compound 48/80 by 31% (Fig. 1, *c*). Casomorphine did not exhibit antioxidant activity in the model system of quercetin autooxidation. It could be suggested that casomorphine potentiates the stimulatory effect of 48/80 on degranulation of MC due to activation of μ-receptors. However, evidence exists that μ-receptor antagonist naloxone does not prevent activation of skin MC induced by opioid codeine [6]. Probably, the effect of the test preparation is mediated by nonspecific mechanisms. Casomorphine probably produces a nonspecific effect. This phenomenon requires further investigations. Casomorphine inhibits ionophore A23187-induced activation of MC, which suggests that this opioid modifies membrane channels involved in Ca^{2+} transport.

Our results show that bioactive compounds differing by chemical structure (amino acid, dipeptide, and polypeptide) produce various effects on functional activity of MC. These data should be taken into account when correcting predisposition to allergic reactions and hypersensitivity of the organism.

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