EFFECT OF N-PHENYLNAPHTHYLENEDIAMINE ON CATECHOLAMINE DISTRIBUTION IN PERITONEAL MAST CELLS

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UDC 612.112.93.018:577.175.52].014,46

KEY WORDS: catecholamines; mast cells; phenylnaphthylenediamine.

Secretion of catecholamines into the blood stream takes place as a result of stimulation of the sympathicoadrenal system and in response to activation of mast cells (MC) [2, 3]. There have been comparatively few luminescence-histolochemical studies aimed at revealing changes in the catecholamine levels in the MC system under the influence of drugs. For instance, after injection of adrenomimetic and adrenolytic drugs changes were found in the catecholamine concentration in the MC system [1, 4].

The aim of this investigation was to study catecholamine levels in peritoneal MC in vitro in response to their stimulation induced by N-phenylnaphthylenediamine (PND).

EXPERIMENTAL METHOD

Experiments were carried out on 60 Wistar albino rats weighing 180-220 g. In the control, physiological saline was added to a culture of peritoneal fluid obtained from 20 animals. In the experiment (40 rats) a 0.1% solution of PND was added to the culture of peritoneal exudate in the proportion of 0.05 mg to 1 ml. A peritoneal suspension was obtained by the method in [4]. Some experiments were undertaken on isolated MC obtained by differential centrifugation in sucrose [4]. Eagle's medium (0.05 ml) was applied to slides stained with a 10% solution of potassium iodate. To it were added 0.05 ml of the peritoneal suspension of MC and the same volume of 0.1% PND solution. The mixture was covered with a coverslip, the edges of which were smeared with petrolatum, and incubated at 37°C for 24 h. The peritoneal fluid was taken from the animals at the same time, allowing for circadian rhythms of biogenic amines. The method in [6] was used for histochemical identification of catecholamines. The specificity of this reaction was monitored by parallel application of a luminescence method [5]. The intensity of luminescence was measured in relative units.

EXPERIMENTAL RESULTS

MC at different levels of differentiation contained catecholamines: juvenile, mature, and functionally active cells (Table 1). The highest content of catecholamines was found in MC treated with PND, evidently due to the ability of the latter to regulate the bioamine level in structures synthesizing biogenic amines and, in particular, in the MC system. MC in a culture of rat peritoneal exudate were large in size and contained numerous granules, which were not very densely distributed, and not infrequently they masked the nucleus partially or totally. MC give a positive chromaffin reaction, i.e., they contain noradrenalin, which stains a golden brown color, the intensity of which is directly proportional to the concentration of the substances (Fig. 1). Three types of MC were identified in films of peritoneal fluid by the use of the Falck–Hillarp method: 1) small cells, regularly circular in shape, with a distinct, bright and confluent greenish yellow luminescence, masking the nucleus; 2) medium sized circular cells with a clearly distinguishable dark nucleus and with separate granules giving greenish luminescence; 3) large, pale orange, degranulating forms of MC.

Under the influence of PND the MC acquired distinct bright green luminescence. Nonluminescent nuclei were clearly visible. Luminescence in the young MC was only a little more intense than in the control and it became pale green and hazy in character; the outlines of the

Department of Human Anatomy and Physiology, K. L. Khetagurov North-Ossetian University, Ordzhonikidze. (Presented by Academician of the Academy of Medical Sciences of the USSR, T. T. Berezov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 103, No. 6, pp. 742-743, June, 1987. Original article submitted June 11, 1986.

0007-4888/87/0006-0847$12.50

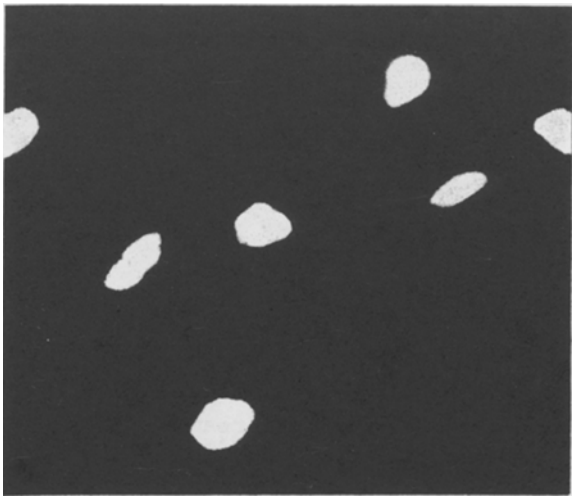

Fig. 1. MC from peritoneal exudate of adult Wistar rat.

TABLE 1. Catecholamine Content (in relative units) in Peritoneal MC after Addition of PND (M ± m)

Type of MC	Control	PND
Juvenile	2,33±0,07	3,20±0,05*
Mature	3,40±0,04	4,69±0,09*
Functionally active	0,80±0,04	0,250±0,06*

Legend. *p < 0.01 compared with normal.

cells were indistinct. The effect of luminescence was particularly marked in mature MC, luminescence in which became bright green in color (Fig. 1). Parallel staining by the Hillarp–Hokfelt method showed that mature luminescent MC gave a positive chromaffin reaction. The stronger the degree of the chromaffin reaction, the more brightly the luminescence of the cell. Up to 22.0 ± 0.6 of them were counted in a field of vision.

On the basis of these results it can be concluded that PND causes an increase in the catecholamine content in MC. This phenomenon can be explained by the following mechanism: PND blocks monoamine oxidase or catechol O-methyl transferase, which metabolize noradrenalin, i.e., it reduces the activity of the system inactivating catecholamines in MC.

LITEATURE CITED

1. G. S. Gordon, V. E. Sergeeva, and I. G. Zelenova, Neurotransmitters of Lymphoid Organs [in Russian], Leningrad (1982), pp. 25-28.
2. P. A. Motavkin and V. S. Korelina, "Localization of biogenic monoamines in mast cells," Arkh. Anat., Gistol. Émbriol., No. 5, 11 (1985).
3. V. E. Sergeeva, The Histotopography of Catecholamines in the Mammalian Thymus [in Russian], Vol. 2, Moscow (1974), No. 4, pp. 1-128.
4. J. C. Padaver and A. S. Gordon, Proc. Soc. Exp. Biol. Med., 88, 29 (1955).
5. B. Falck and N. A. Hillarp, J. Histochem. Cytochem., 10, 348 (1962).
6. A. G. E. Pearse, Histochemistry, Theoretical and Applied, London (1962), p. 805.