### MORPHOLOGY AND PATHOMORPHOLOGY

# VASOPRESSIN IN TISSUE BASOPHILS OF THE ALBINO RAT DURA MATER

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One of the basic conditions determining the role of mast cells in the regulation of vascular tone is that they contain substances with vasoactive properties [1]. In certain cases mast cells are known to cause both constriction and dilatation of blood vessels [6, 8]. Together with other causes, this phenomenon has been explained by the diversity of their chemical effectors, of which the biogenic amines have been studied the most [2].

The aim of this investigation was to identify vasopressin in mast cells of the albino rat dura mater.

#### EXPERIMENTAL METHOD

The dura mater was removed from 11 mature inbred albino rats after decapitation under ether anesthesia. Vasopressin-containing cells were tagged by a two-stage immunohistochemical method [3], using monospecific hog antibodies to arginine-vasopressin (UOCHB CSAV, Czechoslovakia) and rabbit antibodies against hog IgG, conjugated with fluorescein isothiocyanate (FITC) and horseradish peroxidase (HRP) ("Sevac," Czechoslovakia) for luminescence and light microscopy respectively. Tissue basophils were identified after staining with methylene blue [4] by the presence of metachromatic granulation in preparations which had been treated beforehand with antiserum labeled with the fluorochrome for the object used to detect vasopressin. The material was studied in reflected light under the MBI-15-2 luminescence microscope (LOMO) and in transmitted light under the "Ergoval" light-optical microscope (Carl Zeiss, Jena). The number of mast cells was counted in an area of 1 mm<sup>2</sup>. Their mean area also was determined. The results were subjected to statistical analysis.

#### **EXPERIMENTAL RESULTS**

Positive immunoreactivity for vasopressin was found in preparations of the dura mater of albino rats treated with antiserum labeled both with FITC and with HRP. Under the luminescence microscope structures containing vasopressin gave emerald green fluorescence. Their outlines varied from circular, elongated, to irregular in shape. In the center of these cells a round area corresponding to the region of the nucleus, and free from fluorescence, was usually observed (Fig. 1a).

The outlines of cells detected by the HRP method corresponded to those revealed by fluorescence. Their cytoplasm stained brown. A translucent nucleus could be identified in each cell (Fig. 1b). Staining with methylene blue showed that cells with an immunopositive reaction for vasopressin corresponded to mast cells, and the characteristic metachromatic granulation could be seen in their cytoplasm (Fig. 2).

Comparison of the immunohistochemical and morphological pictures showed that the dimensions of all the cells described above were about the same, and did not vary significantly (19.3  $\pm$  1.33); the number of cells stained with methylene blue (14.4  $\pm$  1.3) was almost the same as the number of vasopressin-positive cells revealed by luminescence (14.1  $\pm$  1.11). The number of cells detected by the HRP method was a little less (9.6  $\pm$  0.7), probably because of the lower sensitivity of the method [6].

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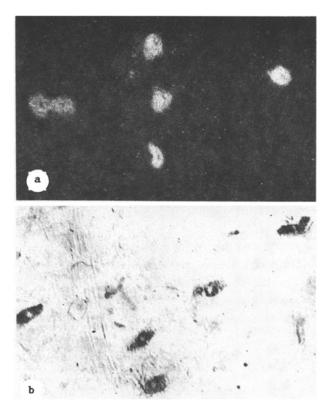


Fig. 1. Localization of vasopressin in mast cells: a) immunofluorescence method, b) immunoperoxidase method. 140×.

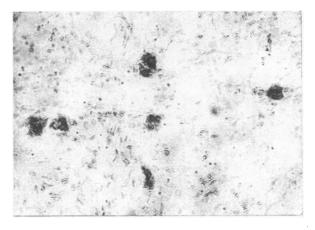


Fig. 2. Mast cells treated with methylene blue. 140×.

Mast cells were distributed irregularly in the dura mater of the albino rats, most frequently in groups along blood vessels. Closeness of mast cells to blood vessels is regarded by most investigators as making possible the realization of the vasoactive action of biogenic amines (serotonin, dopamine, noradrenalin, histamine) contained in their cytoplasm [2]. In addition, the presence of vasoactive intestinal polypeptide (VIP) was demonstrated previously in the tissue basophils of the mucous membrane of the digestive tract [5], and in the present investigation, vasopressin was identified in the tissue basophils of the dura mater.

The results are evidence that mast cells can regulate the cerebral blood flow by secreting, together with biogenic amines, oligopeptide hormones capable of exerting both a modulating and a directly effector action on the monocytes of blood vessels.

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## EFFECT OF TACTIVIN ON THYMUS MORPHOLOGY IN EXPERIMENTAL LOWER LIMB TRAUMA IN MICE (THE ANTISTRESSOR EFFECT OF TACTIVIN)

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In recent years the range of diseases and pathological states accompanied by immune disturbances, for the optimal treatment of which the use of various immunomodulators is indicated, has widened [2]. Changes characteristic of the stress reaction, and due to corticosteroids, develop under these circumstances in the thymus [8, 11]. There are indications in the literature that thymic hormones and, in particular, thymosins can counteract the development of a stress-induced immunodeficiency state [10]. Accordingly, we decided to study the possibility of using tactivin, one of the most active Soviet immunomodulators [1], on a model of experimental trauma, in which a stress-reaction (accidental involution) of the thymus also develops [9], with the aim of speeding up recovery of the organ after trauma.

#### EXPERIMENTAL METHOD

Experiments were carried out on mature female (CBA  $\times$  C57BL/6) $F_1$  mice weighing 17.5-19.0 g. A closed fracture was produced manually in the lower third of the left femur of the experimental mice. Tactivin was injected subcutaneously in a sessional dose of 1.0  $\mu$ g per mouse 1 day before the fracture or immediately thereafter (the mice were killed 24 h after the fracture) or during the 3 days after the fracture (mice were killed 5, 10, and 15 days after the fracture). Animals subjected to similar trauma, but not receiving tactivin, together with intact mice served as the control. Five animals were used at each point. The thymus was removed, weighed, and fixed in Bouin's fluid. Morphometric analysis was carried out on 4- $\mu$ -paraffin sections stained with azure II-eosin, in three different zones: subcapsular, cortical, and medullary. The relative areas occupied by the

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