sociated brain cells in collagen wells are not clear. The fact that characteristic chains are formed more frequently in cultures of dissociated spinal cord cells of 12-14-day mouse embryos suggests that the processes described above may perhaps be dependent on the stage of embryonic differentiation of nervous and glial cells in the spinal cord. It is thus likely that the formation of linear chains of aggregates reflects the organ specificity and histogenetic powers of dissociated nerve tissue cells.

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CIRCADIAN RHYTHM OF MAST CELL FUNCTION IN THE RAT DURA

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KEY WORDS: dura mater; mast cells; biogenic amines.

Processes taking place in the body at the cell, tissue, and organ levels are characterized by a definite rhythm [7]. Mast cells, which contain biogenic amines, are local regulators of tissue homeostasis [5].

The object of the present investigation was to study the diurnal activity of mast cells in the dura mater of rats and changes in their content of biogenic amines during the 24-h period.

EXPERIMENTAL METHOD

The dura of sexually mature noninbred albino rats weighing 180-200 g was studied. The method of Falk and Hillarp was used. The animals were kept under identical conditions and fed twice a day; material was taken every 3 h. The dura, straightened out on slides, was dried at room temperature for 15 min, then treated with gaseous formaldehyde at 80°C for 1 h. The specimens were studied in light in the blue-violet region of the spectrum with a wavelength of 410-480 nm and photographed on highly sensitive RF-2 film. Biogenic amines

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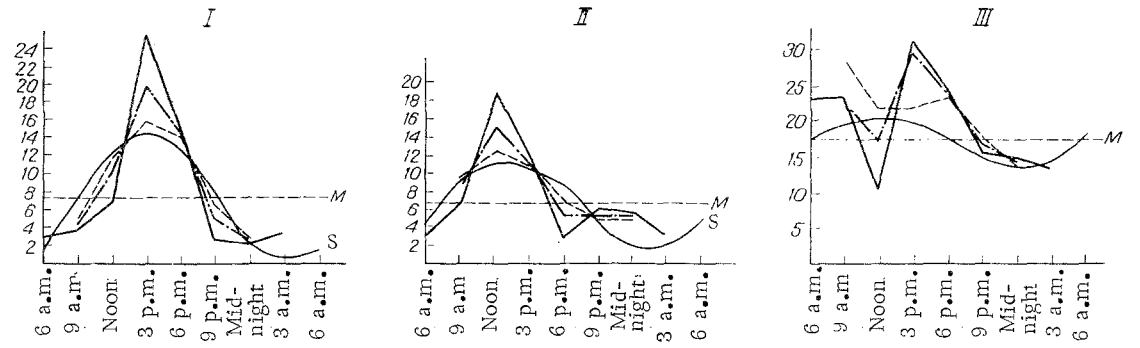

Fig. 1. Changes in number of normal (I) and degranulating (II) mast cells and in content of monoamines (III) in mast cells of rat dura mater at different times of the 24-h period. Continuous line — simple mean, broken line — simple sliding mean, line of dots and dashes — weighted sliding mean. S) Sinusoid, M) mesor.

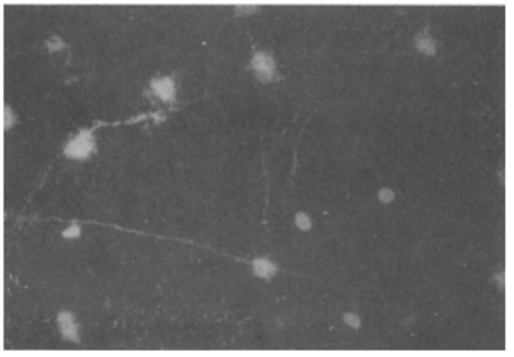

Fig. 2

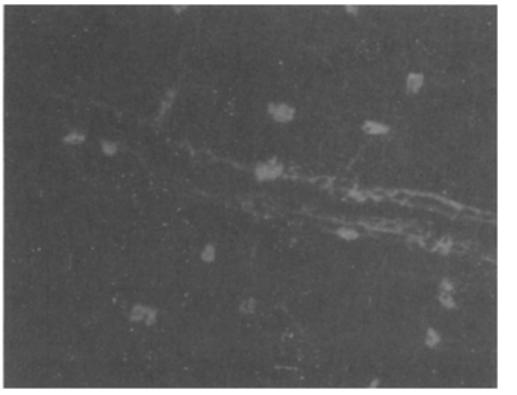

Fig. 3

Fig. 2. Degranulating mast cells of rat dura mater. Falk's method, 200 ×.

Fig. 3. Intact mast cells in rat dura mater. Falk's method, 200 ×.

(serotonin, histamine, catecholamines) in the mast cells were determined quantitatively in total by the FMÉL-1A photometric attachment with a 0.5 mm probe for a single cell.

The mean number of normal mast cells (group measuring up to 10 μ) and of degranulated mast cells (groups measuring from 11 to 20 μ) per square millimeter of surface of the dura and the quantity of biogenic amines in the mast cells at different times of the 24-h period were determined by statistical analysis of the data (Fig. 1). Later, by the method of simple and weighted sliding means, mean values reflecting overall changes in the number of cells tested, largely free from random deviations, were determined [3, 4]. During the study of the rhythm of functional activity of the mass cells a latent rhythm was discovered by determining the duration of the period, the amplitude of fluctuation, the localization of the peak, and the frequency of the rhythm, and also the level about which the system oscillates [1, 2, 6, 7]. The sinusoid plotted as the result of calculations describes periodic changes in the mast cells of the rat dura during the 24-h period.

EXPERIMENTAL RESULTS

Mast cells in the rat dura mater are distributed along the course of the blood vessels and in the substance of the membrane, they differ in size and shape, and give off yellowish-green fluorescence. The bright luminescence of mast cells was observed at 12 noon. At this time degranulation of the mast cells was clearly defined, the cells measured up to 12-14 μ, their outlines were uneven with projecting granules, some of which lay in the intercellular space (Fig. 2). Cells measuring up to 18-20 μ with a dark, nonluminescent nucleus, could be seen in some places. Signs of degranulation of the mast cells were observed at 9 p.m., but activity of extrusion of the granules from these cells was less marked and the diameter of the cells did not exceed 10-12 μ. A sharp decrease in functional activity was observed at

3 p.m. At that time no degranulating mast cells whatsoever could be detected. Most of the cells observed in the tissue were intact mast cells, regularly circular in shape, and measuring 8-10 μ in diameter (Fig. 3). At 6 p.m., and also at night and in the early morning, single mast cells located mainly along the course of the blood vessels could be seen.

Statistical analysis of the data (Fig. 1) showed that the mast cells of the dura mater and the monoamines contained in them are characterized by circadian rhythms with a 24-h period. The characteristics of the sinusoids were found to coincide with those of averaged time curves of the number of normal (Fig. 1, I) and degranulating (Fig. 1, II) mast cells. The sinusoid for the content of monoamines (Fig. 1, III) in the cells differed from the averaged time curves and gives new information on the biological rhythm and, in particular, on the appearance of a maximum. The 24-h sinusoid has stochastic maxima: at 12 noon for degranulating mast cells and at 3 p.m. for normal mast cells and monoamines. The amplitude of the fluctuation is highest for normal mass cells (A = 7.6), and somewhat lower for degranulating cells (A = 3.7). Our observations showed that the acrophase of the monoamine sinusoid occurs sooner than in sinusoids of normal and degranulating mast cells. In the circadian rhythm of normal mast cells and monoamines, besides the 24-h sinusoid it was also possible to distinguish a 12-h sinusoid (6-8 h for monoamines, 9-21 h for cells). The amplitude of the 12-h sinusoid was about half that of the 24-h sinusoid. The 12-h sinusoid may be due to large nervous and energy expenditure, it is more dynamic, it can vary more rapidly, and may weaken considerably in its intensity [5], and it responds more accurately to changes in tissue homeostasis.

The study of the time mechanisms taking place in mast cells of the dura mater thus revealed two functional states of these cells: 1) a peak of degranulating mast cells at 12 noon; 2) a peak of intact mast cells at 3 p.m. These two states of the mast cells correlate with the content of monoamines in them, so that at different moments of time they are able to respond adequately to definite stimuli.

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