

# EXPERIMENTAL BIOLOGY

## CIRCADIAN RHYTHM OF ADRENERGIC NERVE FIBER ACTIVITY IN THE RAT DURA MATER

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KEY WORDS: dura mater; adrenergic fibers; circadian rhythm.

Many biological processes exhibit a circadian rhythm [2, 4]. The sympathetic nervous system changes the state of functional preparedness of organs in accordance with the conditions of existence and plays the role of principal factor for the urgent mobilization of the protective forces of the organism to restore disturbed equilibrium [1].

The object of this investigation was to study diurnal changes in activity of adrenergic nerve fibers of the rat dura mater.

### EXPERIMENTAL METHOD

The dura mater of noninbred mature rats weighing 180-200 g, kept under identical conditions, was studied by the method of Falk and Hillarp. The dura was straightened out on slides and dried at room temperature for 15 min, after which it was treated with gaseous formaldehyde at 80°C for 1 h. The preparations were studied in blue-violet light with a wavelength of 410-480 nm and photographed on highly sensitive RF-2 film. Every hour the mean number of adrenergic nerve fibers in 1 mm<sup>2</sup> of dura was counted. During subsequent statistical analysis the values of the simple and weighted sliding means and also the duration of the period, amplitude of oscillations, and frequency of the rhythm were calculated. A sinusoid (Fig. 1) was plotted from the results of the calculations (Fig. 1). The parameters of the sinusoid were determined by the method of least squares [3, 5, 6]. A cosinusoidal function was used for approximation.

### EXPERIMENTAL RESULTS

Adrenergic nerve fibers form a plexus in the dura mater consisting of nerve trunks and single nerve fibers, arranged mainly along the course of blood vessels. On arterioles of the

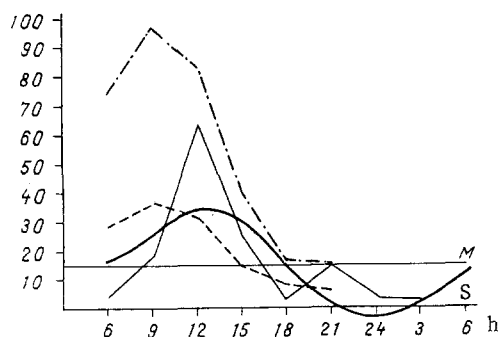


Fig. 1. Changes in number of adrenergic nerve fibers in rat dura mater during the 24-h period. Continuous line — simple mean; broken line — simple sliding mean; dot-dash line — weighted sliding mean. S) Sinusoid; M) mesor.

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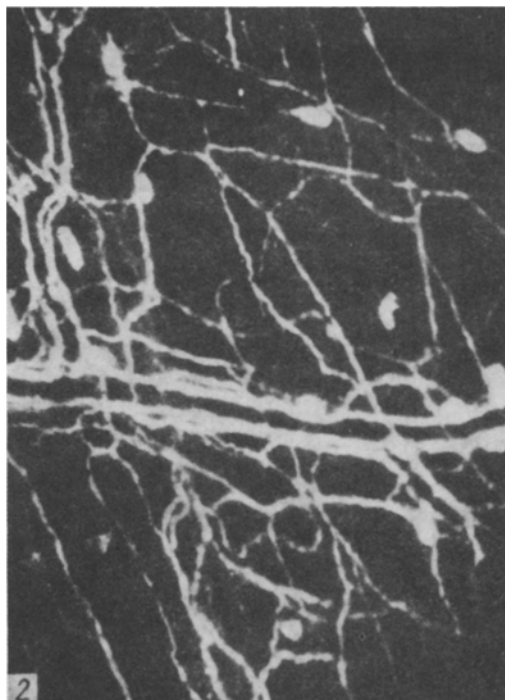


Fig. 2

Fig. 2. Adrenergic nerve fibers in rat dura mater at 12 noon. Falk's method, 200 $\times$ .

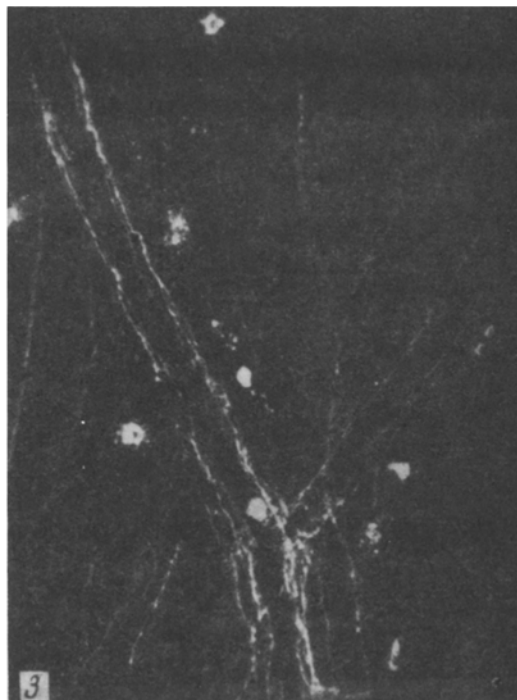


Fig. 3

Fig. 3. Adrenergic nerve fibers of rat dura mater at 6 p.m. Falk's method, 200 $\times$ .

deep layer of the dura mater they form very thick nerve plexuses. Single varicose fibers could be seen on the walls of all visible blood vessels, probably including the smallest, terminal arterioles. Luminescence of the nerve fibers at 12 noon was bright. At that time the adrenergic fibers gave fluorescence of a saturated green color (Fig. 2). The adrenergic fibers became varicose, their branches were interwoven, and the degree of branching varied. Single branches were observed in the tissue of the dura but along the course of the vessels the fibers were grouped in bundles. A sharp fall in activity was observed at 6 p.m. (Fig. 3). At that time the fibers were pale, with no varicosities, and separate nerve fibers could be observed in the tissue of the dura and along the course of the blood vessels.

Statistical analysis of the data showed that adrenergic nerve fibers exhibit a circadian rhythm with a period of 24 h. The rhythm has two maxima: At 12 noon and 9 p.m., and the number of adrenergic fibers is greatest at 12 noon; the increase in their number at 9 p.m. is small. Since this additional peak at 9 p.m. does not reach the level of statistical significance ( $P = 0.05$ ), its independent existence can be disregarded, and the circadian curve of activity of the adrenergic nerve fibers can be regarded as monomodal.

The difference in character and density of the adrenergic innervation of the dura in the course of the 24-h period determine the differences in type of adrenergic function, which varies from general, or unlocalized, with slow regulation of the effector tissue, to specific, accurately localized, and fast [1].

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# HOMO- AND HETEROSPECIFIC TRANSPLANTATION OF EMBRYONIC NERVE TISSUE

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KEY WORDS: heterospecific transplantation; differentiation; neurons.

Attempts to transplant embryonic nerve tissue into the brain of an adult animal of the same species were made several decades ago [1, 6, 8]. It was then shown by morphological methods that the grafts may take and differentiate in the recipient's brain. A long time after these investigations the second stage began: investigation of grafts with the combined use of morphological, histological, and autoradiographic methods [2, 3, 7, 9, 11, 12]. The results of these investigations showed that blood vessels which provide for normal nutrition grow into the graft, neurons differentiate from neuroblasts, and the architectonics of the structure which was transplanted is formed, with preservation of its biochemical properties. For example, grafts of the raphe nuclei synthesized serotonin, grafts of the septum synthesized acetylcholine [2, 3]. Fibers from the graft can grow toward neighboring structures of the recipient's brain and can also receive afferent connections from them [4, 10, 12].

These facts suggest that the function of injured structures of the recipient's brain can be restored and replaced by a graft. Despite obvious specific difficulties, transplantation into the brain does not meet with the main fundamental difficulty, the problem of rejection of foreign tissue through the action of immune mechanisms. The presence of the blood-brain barrier in transplantation into the brain theoretically allows transplantation of the tissue not only of another individual, but also of another biological species. The object of the present investigation was to make a comparative experimental study of homo- and heterospecific transplantation of nerve tissue.

## EXPERIMENTAL METHOD

The operation was performed under sterile conditions. The recipient animals (rats and rabbits) were scalped under pentobarbital anesthesia (40-50 mg/kg body weight) and fixed in SEZh-2 or STM-3 stereotaxic apparatuses in accordance with coordinates taken from the atlas of Fikova and Marsale [5]. A hole 5 mm in diameter, with coordinates of its center AP = 0, L = 0.5 mm for rabbits and AP = +1, L = 0.5 mm for rats, was then made in the cranial bones

TABLE 1. Transplantation of Embryonic Rat Septal Tissue into Brain of Rats and Rabbits

Recipient rats			Recipient rabbits		
No. of animal	duration of expt., days	graft	No. of animal	duration of expt., days	graft
1*	50	Glia	1	150	Glia
2	60	"	2	30	Neurons
3*	40	Neurons	3	25	"
4	40	"	4	40	"
5	180	No graft	5	45	Glia
6*	190	Neurons	6	60	No graft
7	190	No graft	7	120	Neurons
8	200	Neurons			
9	200	"			

\*Noninbred rats were used as recipients.

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