DISTURBANCE OF SPERMATOGENESIS IN RATS WITH CHRONIC EMOTIONAL STRESS

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Disturbance of normal reproductive processes is often observed in animals with emotional stress [1, 4, 14]. There is abundant evidence in the literature on the effect of emotional stress on stability of the estrous cycle [11, 13], and gonadrotropin and sex hormone levels [2, 8, 10]. It has been shown that zoosocial stress is accompanied by a decrease in weight of the testes and of the accessory sex glands [6], and by reduction of the size of the Leydig's cells, whose diameter decreases to 20% of the initial value [9]. However, the consequences of emotional stress directly on differentiation of the sex cells in the testes have not been adequately studied.

The aim of this investigation was to study the effect of chronic emotional stress on spermatogenesis in rats.

EXPERIMENTAL METHOD

Experiments were carried out on 12 male Wistar rats weighing 250-300 g. To create emotional stress the animals were fixed in plastic containers and subjected to stochastic electrodermal stimulation through subcutaneous needle electrodes, fixed to the spine and at the base of the tail, with a square positive pulses. The parameters of stimulation were: amplitude 5 V, frequency 100 Hz, pulse duration 10 msec, duration of series of pulses from 30 to 60 sec. The animals were stressed 3 h daily for 7 days. After the end of the experiment the animals were decapitated and the thymus, adrenals, seminal vesicles, and testes were removed. The glands were weighed. The testes were fixed in Carnoy's fluid and stained with hematoxylin and eosin. Paraffin sections from 2 to 5 μ m thick were examined under the light microscope. The number of "plugs" — conglomerates of degenerated sex cells — was counted in 100 randomly chosen sections of the convoluted seminiferous tubules. The morphometric method of counting the index of spematogenesis also was used in the analysis [5]: the degree of differentiation of the spermatogenic layer was estimated by a 4-point system in 100 randomly chosen sections through the convoluted seminiferous tubules and the averaged index was calculated. By means of this method it was possible to determine the general level of preservation of the spermatogenic layer in a given animal. The results were subjected to statistical analysis by Student's test.

EXPERIMENTAL RESULTS

As a result of emotional stress for 7 days the animals were found to develop hypertrophy of the adrenals and involution of the thymus, characteristic features of the classical stress syndrome as described by Selye [3]. A significant increase in mass of the adrenals and a decrease in mass of the thymus was accompanied by a tendency toward a decrease in mass of the testes and seminal vesicles compared with the control animals (Table 1).

The results of light microscopy showed a significant increase in the number of "plugs" in the convoluted seminiferous tubules of the stressed rat compared with the controls. Whereas the number of "plugs" in the control was $19.9 \pm 5.5\%$, after stress it was 55.2 ± 4.4 (p < 0.01). The index of spermatogenesis in the experimental animals was significantly lower than in the

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TABLE 1. Effect of Chronic Emotional Stress of Mass of Gland (in mg)

Gland	Control (n = 6)	Stress (n = 6)
Adrenal Thymus Testis Seminal vesicle	$\begin{array}{c} 48.5 \pm 3.8 \\ 486.0 \pm 50.5 \\ 3000.0 \pm 130.0 \\ 1066.7 \pm 91.9 \end{array}$	$59.7 \pm 3.3*$ $305.8 \pm 47.5*$ 2800.0 ± 60.0 883.3 ± 132.7

Legend. *p < 0.05 compared with control.

controls. The index of spermatogenesis of the stressed rats 3.38 ± 0.03 compared with 3.56 ± 0.02 (p < 0.01) in the controls. Microscopic investigation of the experimental animals revealed solitary regions of convoluted seminiferous tubules, in which the sex cells were represented only by spermatogonia and spermatocytes, by contrast with the controls. This may indicate that during emotional stress lasting one week injury to the spermatogenic cells takes place primarily at the stage of spermatogenesis.

Thus, chronic emotional stress, leading to typical changes in the adrenals and thymus leads at the same time to marked changes in processes of spermatogenesis. The increase in the number of "plugs" in the convoluted seminiferous tubules is evidence of the appearance of regions with a damaged spermatogenic layer in the testes. It can be postulated that the reduction of the index of spermatogensis against the background of emotional stress takes place as a result of the more rapid elimination of the sex cells. Thus, on microscopy an increase in the number of regions without spermatocytes was observed. The results are in agreement with those of an investigation [7] in which inhibition of spermatogensis was observed in white-footed mice, manifested as disappearance of spermatids and an increase in the number of the preceding embryonic elements.

Disturbances of spermatogenesis which we found can be explained by a decrease in concentrations of FSH and testosterone, under the control of which the sex cells develop at the stages of spermatocytes and spermatids [14]. Reduction of the weight of the seminal vesicles, the target of action of the hormone testosterone [12], as a result of 7 days of emotional stress is evidence of a probable decrease in the plasma concentration of this hormone, and this could lead to the development of the disturbances of spermatogenesis discoverfed in rats during chronic emotional stress.

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