MITOTIC ACTIVITY OF MAMMARY GLAND TISSUES AFTER TRAUMA IN NONCASTRATED AND CASTRATED RATS

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Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 56, No. 10,
pp. 89-92, October, 1963
Original article submitted January 8, 1963

The mitotic activity of the epithelium of the mammary gland has been studied mainly in animals in various hormonal states [1, 4, 5, 6]. It was studied by I. V. Markelova [7] during posttraumatic regeneration, and her results showed that in albino mice (sexually mature females in the stage of diestrus), after removal of 50-70% of the total mass of the mammary gland tissue, the mitotic coefficient of the residual part of the organ is not appreciably altered on the 6th-15th day after the operation. Isolated items of information concerning mitotic division of cells in mammary gland tissue after injury to the organ are given by V. P. Vlasov [2], D. I. Golovin [3], and Coen [8]. These authors, however, did not actually count mitoses and did not take into account the stage of the estrous cycle in their animals. We could find no information in the literature concerning the mitotic activity of the mammary gland tissues after trauma in castrated animals.

EXPERIMENTAL METHOD

The investigation was conducted on parous albino rats (44 noncastrated and 37 castrated). Castration was performed under general ether anesthesia in aseptic conditions. The animals were used in the experiment 20 days after the operation. A deep thermal burn of the tissues of the right abdomino-inguinal mammary gland was inflicted by means of a red-hot disk measuring 0.2 × 0.4 cm. The rats were sacrificed at the same time of day (10-11 A.M.) 1, 3, 5, 7, 10, 15, 20, 30, 40, and 60 days after injury. Several noncastrated and castrated rats without injury to the mammary gland were also used.

Material for cutting sections was fixed in Carnoy's and Brank's solutions and in Zenker formol. Paraffin-wax sections, cut to a thickness of 5-7 μ, were stained with Heidenhain's hematoxylin, azure-II-eosin, and by Feulgen's method. The stage of the estrous cycle was determined in all the animals from the cell picture in vaginal smears. Total preparations, fixed in formalin and stained with hemalum, were also made from the theoracic mammary glands which were flattened in shape; these preparations were used to determine the animal's physiological state [9-11].

All the castrated rats were in the stage of diestrus. The overwhelming majority of noncastrated rats were also in a stage of diestrus, but a few animals were in other stages of the estrous cycle. We therefore give the numerical data relating to mitotic activity for all the noncastrated animals separately, and only for the animals in the stage of diestrus. On the basis of these figures the mitotic coefficient per 1000 cells was calculated. The results were analyzed statistically by the Fisher-Student method.

EXPERIMENTAL RESULTS

Since the study of reparative regeneration of the mammary gland has been conducted on guinea pigs, dogs, rabbits, and mice but not on rats a short account of the process of regeneration in the latter is given below.

Twenty four hours after injury reactive changes (an increase in the number of cells) begin to develop in the glandular tissue around the zone of necrosis. The number of mitoses is not visible increased. However, inflammatory manifestations are already well marked (Fig. 1). After 3 days the number of mitoses in the epithelium of the lacteals and alveoli and also in the endothelial cells of the blood vessels is increased (Fig. 2). After 5-7 days dedifferentiation of the lacteals and alveoli, on the one hand, and development of granulation tissue, on the other hand, are seen. The

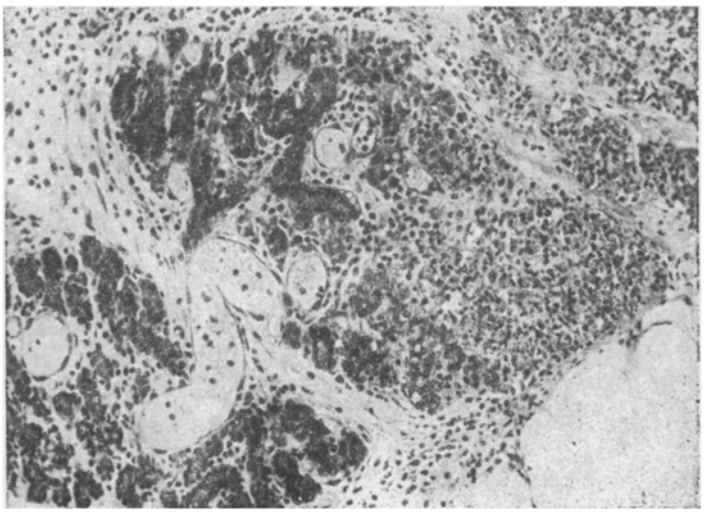

Fig. 1. Mammary gland tissue of a noncastrated parous rat close to the site of injury 24 h after infliction of a burn. Dilated blood vessels, leukocytic inflammatory infiltration. Photomicrograph. Fixation in Brank's solution. Heidenhain's hematoxylin. Objective, apochromatic 20 ×, homal II.

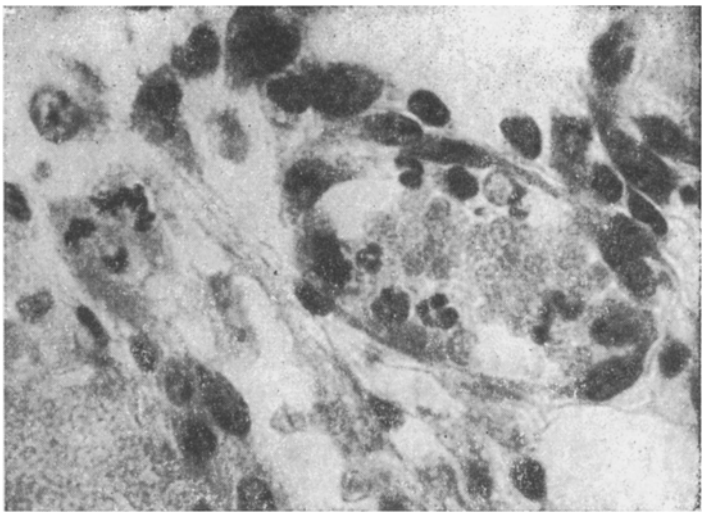

Fig. 2. Three mitoses in endothelium of blood vessels of the mammary gland of a noncastrated parous rat 3 days after burning. Photomicrograph. Fixation in Carnoy's solution. Azure-II-eosin. Objective, apochromatic 60 ×, ocular 10 ×.

mitotic activity of the cells of the epithelium and granulation tissue under these conditions showed a sharp increase in both the noncastrated and the castrated rats, reaching a maximum after 7 days. The process of mitotic division of the cells continued at subsequent periods; new lacteals were formed as a result of division of the epithelial cells.

It must be emphasized that in every case different phases of mitosis were observed in both epithelium and connective tissue. This is easily seen in Fig. 3: 10 days after injury 3 mitotically dividing epithelial cells may be seen in one of the terminal portions of the gland in a castrated rat – two of these are in the metaphase and one in the anaphase.

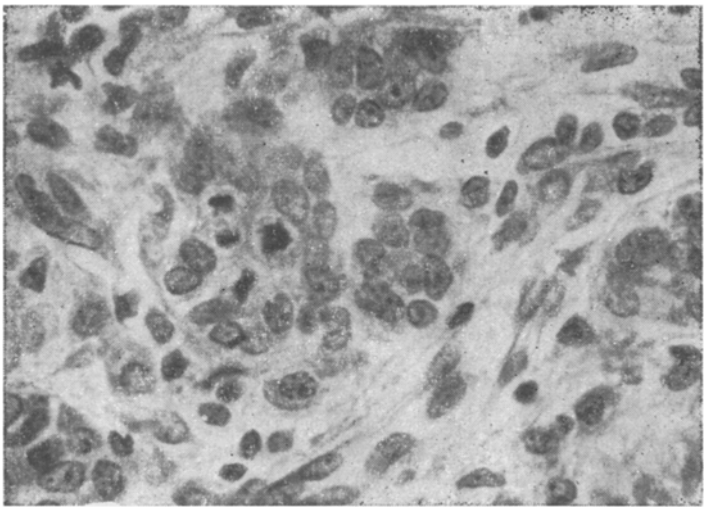

Fig. 3. Three mitoses in the alveolus of a mammary gland of a parous castrated rat 30 days after castration and 10 days after burning. Photomicrograph. Fixation in Brank's solution. Heidenhain's hematoxylin. Objective, apochromatic 60 ×, homal VI.

Changes in the Mitotic Coefficient of Tissue Cells of the Mammary Gland during Postraumatic Regeneration in Noncastrated and Castrated Rats

Rats	Time after burning (in days)	Noncastrated rats					Castrated rats		
		Number of rats	Mitotic coefficient				Number of rats	Mitotic coefficient	
			All rats		Rats in a stage of diestrus				
			epithelium	connective tissue	epithelium	connective tissue		epithelium	connective tissue
Control		5	0.5	0.2	0.5	0.2	5	0.1	0.2
Experimental	1	3	0.5	0.2	0.3	0.2	2	0.1	0.2
	3	2	0.7	2.7	0.7	2.0	2	0.7	2.3
	5	3	1.4	3.3	1.4	3.3	2	0.9	2.9
	7	4	2.8	4.4	2.7	4.3	4	1.7	4.2
	10	3	1.9	2.5	1.9	2.5	4	1.2	2.4
	15	5	0.2	0.2	0.2	0.2	3	0.2	0.2

After 15 days the necrotic masses in the focus of injury were completely absorbed and in their place granulation tissue developed. This was invaded by undifferentiated epithelial bands and tubes from areas of grandular tissue previously undergoing changes. The mitotic activity of the dividing cells at this stage was much reduced and was nearer to that observed in normal conditions.

The numerical data characterizing the activity of mitotic division of the epithelial and connective-tissue cells during posttraumatic regeneration are given in the table, which shows that the mitotic activity of the cells of the interstitial connective tissue in the intact mammary gland of the noncastrated and castrated rats was identical. In the epithelium, on the other hand, the mitotic activity in the noncastrated rats was five times greater than in the castrated animals. It must be emphasized that although the cells undergoing mitotic division in the epithelium of the castrated rats accounted for only a very small percentage of the total number of cells, they were found regularly (in 7-10 of the sections from 3 of the 5 animals on which counts were made).

After injury, the number of mitoses in the mammary gland increased in accordance with a regular pattern, reaching its highest value after 5-7 days. The dynamics of the mitotic activity of the connective-tissue and granulation-tissue cells in the noncastrated and castrated rats was absolutely identical as regards both time and number (this is

particularly clear from a comparison of the findings obtained in castrated rats and noncastrated rats in a state of diestrus). On the other hand, the mitotic activity of the epithelial cells 5, 7, and 10 days after trauma was slightly higher in the noncastrated animals.

Statistical analysis of the results shows that the difference between the noncastrated and castrated control rats is significant ($P < 0.05$). The difference between the noncastrated and castrated experimental rats is not significant ($P > 0.05$).

SUMMARY

Dynamics of mitotic activity of the tissue cells of the mammary gland was studied in the process of posttraumatic regeneration in the parous albino rats (44 noncastrated and 37 castrated ones). These experimental studies demonstrated that the influence of ovarian hormones, which had a distinct effect on the mitotic regime of the epithelial cells in the uninjured mammary gland, became of secondary importance in the course of the changes developing after its injury. Local tissue are prominent here.

LITERATURE CITED

1. A. D. Al'tman, Byull. éksper. biol. 20, 10-11, 47 (1945).
2. V. P. Vlasov, Regeneration of the mammary gland and the influence of fibrolysin and iodine on this process. Doctorate dissertation, St. Petersburg, 1909.
3. D. I. Golovin, Trudy Kishinevsk. med. inst. 5, 131 (1956).
4. S. S. Laguchev, Byull. éksper. biol., 9, 108 (1958).
5. S. S. Lagushev, in book: Problems in Regeneration and Cell Division (Proceedings of a Conference on Regeneration and Cell Proliferation) [in Russian], Moscow (1959), p. 250.
6. S. S. Laguchev, Byull. éksper. biol., 3, 83 (1962).
7. I. V. Markelova, Byull. éksper. biol., 12, 84 (1962).
8. E. Coen, Beiträge zur normalen und pathologischen Histologie der Milchdrüse. Jena (1887).
9. H. A. Cole, Proc. roy. Soc. B., Vol. 114, (1933), p. 136.
10. A. J. Dalton, in book: A. Symposium on Mammary Tumors in Mice. Washington, (1945), p. 39.
11. C. W. B. Turner, in book: E. Allen, Sex and internal Secretions. Baltimore, (1934), p. 544.

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
