THE EFFECT OF REPARATIVE REGENERATION OF THE LIVER ON THE MITOTIC ACTIVITY OF THE CORNEAL AND EPIDERMAL EPITHELIUM IN MICE

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Changes in the intensity of cell proliferation in the tissues arising after trauma are not confined to the site of injury. For instance, after injury to the liver or extirpation of part of the organ, considerable intensification of cell proliferation takes place not only close to the wound surface but also in other lobes of the liver not affected by the injury [1, 7]. If a burn is inflicted on one cornea in rats, changes take place in the number of mitoses in the intact cornea of the opposite eye [6]. The regular changes in mitotic activity in the epidermis of rats arising after infliction of a skin wound are accompanied by changes of a similar nature in the number of mitoses in symmetrically opposite areas of intact epidermis [5]. The regenerative processes arising as a result of trauma and reflected in the mitotic activity of the tissues thus affect the intensity of physiological regeneration of injured tissues. M. A. Vorontsova and L. D. Liozner [2] have developed the view that the recovery processes in reparative regeneration are based on the power of the tissues to undergo physiological regeneration, and stress that very little research has been done into the problem of establishing a relationship between these processes. This makes it difficult to analyze the relationship between the restorative processes in physiological and reparative regeneration. The problem of how the level and character, for example the diurnal rhythm, of mitotic activity varies in different tissues when a focus of reparative regeneration is present in the body is another which has been insufficiently investigated.

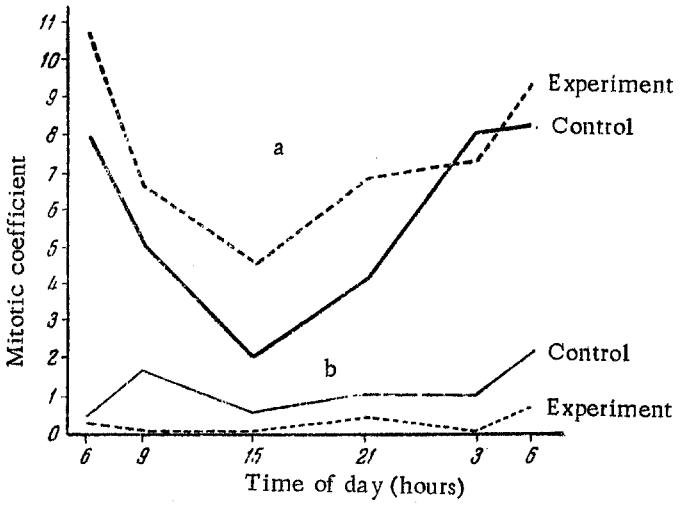
The aim of the present work was to study the changes in the mitotic activity of the epithelium of the cornea and epidermis in mice after extensive injury to the liver.

METHOD

The experiments were conducted on white mice, from which the left lateral and central lobes of the liver (60-70% of the organ) had been removed. The animals were sacrificed by decapitation on the second and third days after operation, at 6 A.M., 9 A.M., 3 P.M., 3 A.M., and 6 A. M. At each of these times 5 experimental and 5 control animals were studied.*

The material for histological examination was fixed in Zenker's fluid. Total preparations were made from the corneas of both eyes, and stained with hematoxylin by Carazzi's method. The concha of the external ear, cut longitudinally from base to apex, was embedded in celloidin-paraffin wax, and sections were cut to a

*The work was carried out on the same animals as were used for the investigations by L. D. Liozner, Z. A. Ryabinina and V. F. Sidorova [7] into the pattern of the mitotic activity of the liver during its reparative regeneration.



Changes in the mitotic coefficient in the corneal epithelium (a) and in the epidermis (b) of the experimental and control mice.

thickness of 8μ. These sections were also stained with Carazzi's hematoxylin. All the sections were examined under an immersion objective 90× and an ocular 7×.

The mitoses in the cornea were counted on both sides of two mutually perpendicular lines drawn on the slide and intersecting at the center of the cornea. In addition to the mitoses in various areas of the cornea, in certain fields of vision of the microscope we counted all the cells in order to calculate their mean number per field of vision. In each cornea the mitoses were counted in 120-140 fields of vision.

The mitoses and cells in the epidermis of the ear were counted on both the external and internal surface of the conchae in 40 fields of vision, on each side, starting from the tip of the auricle. On the average there were 7000-9000 cells in each case.

For both the epidermis and the corneal epithelium we calculated the mitotic coefficient – the number of mitoses found per 1000 cells. The figures obtained were treated statistically by the Fisher-Student method.

RESULTS

Corneal epithelium. The results of the counting of the mitoses in the corneal epithelium of the experimental and control animals are shown graphically in the figure. The curves show that the mitotic activity in the corneal epithelium of the animals of both groups underwent regular changes in the course of the 24 hours. The highest mitotic activity was observed at 6 A.M., after which it gradually fell and, nine hours later, i.e., at 3 P.M., it reached its minimum. This fall in the number of mitoses in the time interval between 6 A.M. and 3 P.M. was statistically significant (for the group of control animals $P = 0.001$ and for the experimental group $P = 0.01$). The mitotic activity then gradually increased. In the group of control animals it reached its maximum at 3 A.M. and remained at this level until 6 A.M. The increase in the mitotic activity in the control animals in the time interval from 3 P.M. to 3 A.M. was statistically significant ($P = 0.004$). In the group of experimental mice the gradual increase in the number of mitoses continued until 6 A.M. ($P = 0.01$).

The figures obtained, showing the diurnal variation in the number of mitoses in the corneal epithelium of mice, were in full agreement with the results of M. T. Gololobova's investigations [3, 4], apart from the fact that in our investigations the minimum number of mitoses occurred at earlier hours than was found in Gololobova's experiments.

Comparison of the course of the curves of variation of mitotic activity in the control and experimental mice shows that the mean values of the mitotic coefficient in the experimental animals at most times of day were higher than in the controls. At 3 P.M. this difference was statistically significant ($P = 0.01$) and at 9 P.M. it was close to significant ($P = 0.05$).

It may hence be concluded that reparative processes in the liver have a stimulating effect on the intensity of physiological regeneration in the corneal epithelium in mice.

Besides counting the mitoses in the cornea of the experimental and control mice, we determined the phases of mitotic division of the cells. We were unable to detect any significant changes in the relative proportions of the individual phases of mitotic division at any time of day. The ratio of early to late phases of mitosis in the control and experimental mice was close to unity.

Epidermis. The changes in mitotic activity found in the epidermis of the external ear in the normal and experimental mice are shown graphically in the figure.

In the first place attention must be drawn to the relatively low level of mitotic activity in this tissue. This factor was obviously the reason why the diurnal rhythm of mitosis in the epidermis was less clearly marked than in the cornea. In the epidermis of normal mice at 6 A.M. a small number of mitoses was found. At 9 A.M., however, their number had risen considerably and had reached its maximum level. Such an increase in mitotic activity during this interval of time was close to significant ($P = 0.02$). The number of mitoses then fell again, reaching its minimum value at 3 P.M. ($P = 0.05$). In the interval between 3 P.M. and 3 A.M., the number of mitoses increased very slowly, but in the interval from 3 A.M. to 6 A.M. the mitotic activity increased considerably ($P = 0.05$). Comparison of the values of the minimum mitotic activity observed at 3 P.M. with the mitotic activity at 6 A.M. next morning shows that this increase was statistically significant ($P = 0.01$).

The figures obtained, showing the change in the number of mitoses in the epidermis over a period of 24 hours, were close to those obtained by M. T. Gololobova [3, 4] in experiments with the epidermis of rats and mice. In her investigations, however, a higher level of mitotic activity of the epidermal cells was found.

The curve reflecting the diurnal variations in the number of mitoses in the epidermis of the experimental mice in its general form repeats the course of the curve of the changes in the number of mitoses in the control animals. Because of the very low values of the mitotic coefficient, however, it is not possible to show statistically whether the differences in the number of mitoses at the different times of day were significant.

During the comparison of the number of mitoses in the epidermis of the control and the experimental mice, attention is primarily drawn to the fact that at all times of the investigation the mean mitotic activity in the experimental animals was lower than the mean mitotic activity in the control mice. These differences, especially in the period of high mitotic activity in the epidermis of the control mice, were statistically significant. For instance, at 9 A.M. $P = 0.001$, at 3 A.M. $P = 0.01$ and at 6 A.M. on the third day $P = 0.01$.

It may be concluded from these findings that extensive injury to the liver and the ensuing repair processes in the organ lead to a lowering of the level of mitotic activity of the epidermal cells of experimental mice.

If the results obtained in this work, on the changes in the mitotic activity of the corneal epithelium and the epidermis of the external ear after the operation of partial hepatectomy, are analyzed it may be concluded that different tissues react differently to injury of the liver: in the cornea of mice an increase in mitotic activity may be observed after operation on the liver, but in the epidermis, on the other hand, depression of mitotic activity takes place. The actual causes of the differences in the response reaction of these tissues cannot yet be established without special investigations.

SUMMARY

The author studied changes in the mitotic activity in the epithelium of the cornea and in the epidermis of the conchae auriculae on the 2nd and 3rd day following reaction of 2/3 of the liver. It was found that the mitotic activity in the epithelium of the cornea in the experimental animals was much higher than in the control group. Diurnal variations in the mitotic activity typical of normal animals was retained. Mitotic activity in the epidermis of the ear in the experimental group of animals was at all times lower, on the average, than in the control group.

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