INJURY AND RESTORATION OF STRUCTURE IN INTERNEURONIC SYNAPSES FOLLOWING TOTAL-BODY X-RAY EXPOSURE

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In the morphological works devoted to the study of the reactions of the nervous system to the action of penetrating radiation, the state of the interneuronic synapses in these conditions has received little study. We know at present that synapses are altered after irradiation [5,6,7], but in the conducted works the state of the synapses in relation to the dose and the time elapsing after the action of ionizing radiation was not investigated. The restoration of the synapses in animals surviving after exposure has not been studied either.

Our aim in this work was to discover the initial changes in the state of synapses due to a single exposure of animals to small, medium, and large doses of x rays, and to trace these changes to the climax of the process and after its completion.

Experimental Method

We studied the synapses in the superior cervical sympathetic ganglia of cats irradiated on the RUM-11 apparatus with doses 30, 100, 500, and 1200 r, at voltage 200 kv, current 15 mA, no filters, and at a distance of 65 cm from the anticathode. The dose rate was about 16 r/min.

In order to check the state of the animals, we made analysis of their blood before irradiation and at different times after irradiation.

The experimental cats (48 animals) were divided into four equal groups. The animals in each group were irradiated with a single dose and sacrificed (in pairs) at different times after irradiation (see table).

To reveal the synapses in the ganglia, we used the Bielschowsky-Gros-Lawrentjew and the Golgi-Deineka methods. In order to compare these methods, we treated the left ganglia by the first method and the right ganglia by the second method in every case. Immediately after the animals were chloroformed we extracted the ganglia and fixed them for $1\frac{1}{2}$ hr in AFA fluid. We then transferred the left ganglia to 20% neutral formalin for subsequent treatment by the Bielschowsky-Gros-Lawrentjew method, and the right ganglion to a 3% silver nitrate solution for study by the Golgi-Deineka method.

Experimental Results

We were able to establish the normal structure of the synapses in the superior cervical ganglion of the cat from our own observations [2,3] and the information in the literature [4,5,7].

Time of Taking Material After Irradiation of Animals

Experimental series	Dose (in r)	Hrs	Days												
		18	1	2	3	4	5	6	7	8	9	16	30	60	80
First	30		X		X				X		X	X		X	
Second	100						X		X	X	X	X	X		
Third	500						X	X	X		X		X		X
Fourth	1200	X		X	X	X	X				X				

Symbol: X – animal sacrificed to obtain material

The synapses in the investigated ganglion consisted of two types – terminal and transitory. The first have the form of rings or bulbs situated at the end of the terminal threads of the preganglionic fibers. The second consist of fine fibrils with thickenings or rings along their length, which contact the bodies and dendrites of the ganglionic neurones. In preparations obtained by the Bielschowsky-Gros-Lawrentjew method, the synapses were stained a pale brown color, and their average diameter was 3μ. The rings were extremely delicate with smooth, distinct outlines.

A study of the preparations of the first series of the experiment (dose 30 r) showed that there were no deviations from normal in the structure of the synapses in any of the animals at any period of the experiment. The first, slight changes in the synapses did not appear until the second series of experiments, where the animals were exposed to a dose of 100 r. On the 9th day after exposure, preparations made by the Bielschowsky-Gros-Lawrentjew method revealed argentophilia in some of the synapses. In preparations obtained by the Golgi-Deineka method these changes were not discovered, since all the synapses had a brownish-black color. Thus, though the latter method demonstrates the synapses more completely and more reliably, it cannot be used for revealing the initial changes in the nerve endings.

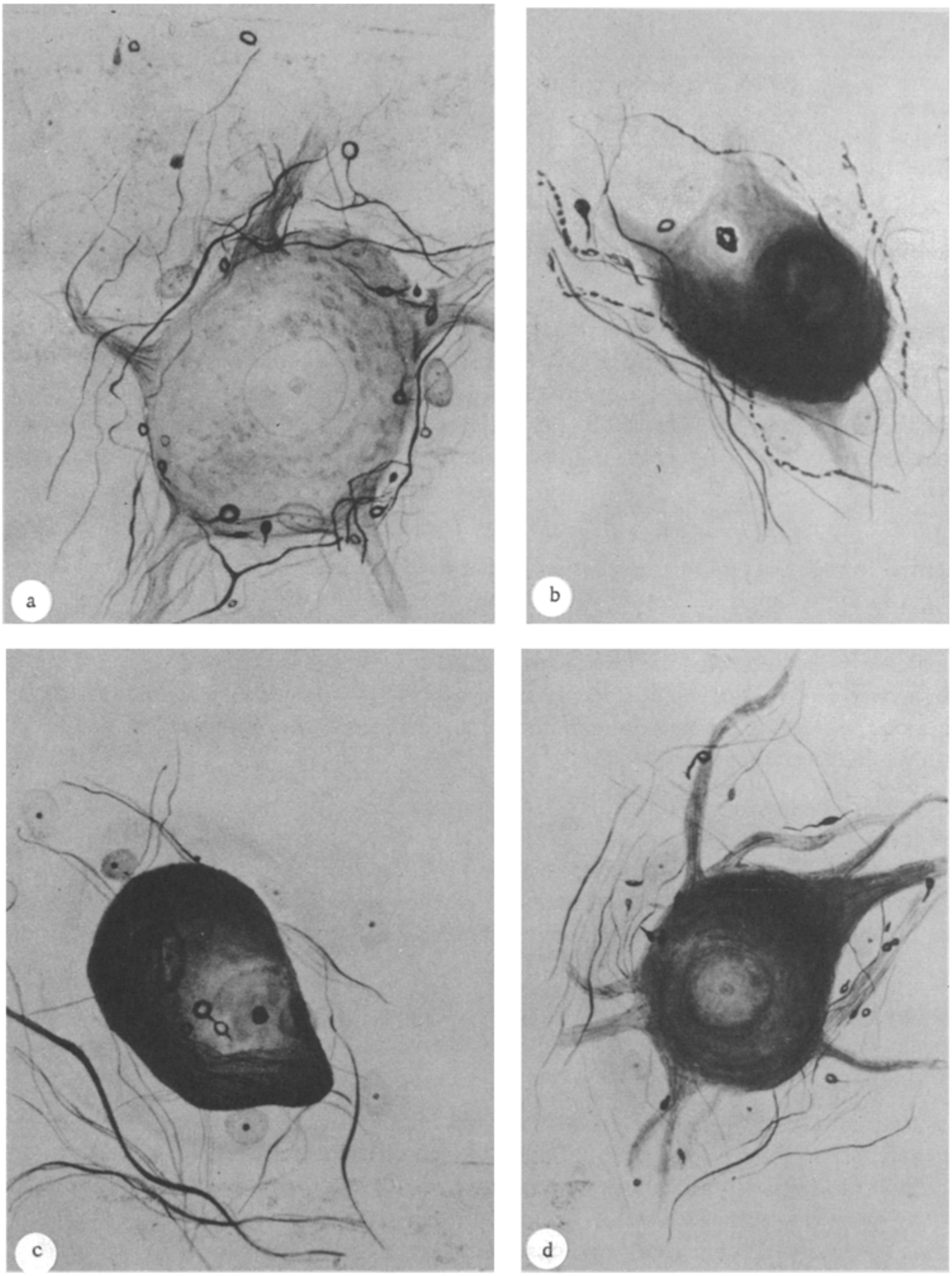
In the third series of experiments (dose 500 r), the animals developed radiation sickness, which was manifested in leucopenia, loss of weight, falling hair, etc. In this series we succeeded in detecting changes in the synapses by the 7th day after exposure to ionizing radiation (see figure, a). In addition to the changes which we observed in the previous series, there was also enhanced silver impregnation of the fine branches of the preganglionic fibers, some of which had a twisted form. In view of this, such fibers could be clearly distinguished from the processes of the ganglionic cells.

On the 9th day after exposure, most of the synapses had the form of darkly stained thickened rings and bulbs. Deformation and increase in size up to 5-8μ were observed. Some of the fibers of the intercellular plexus had varicose swellings and were fragmented in places.

The greatest changes in the synapses were found in the fourth series (dose 1200 r). The first signs of the reaction of the observable structures were manifested on the 5th day after exposure. These initial changes were similar to those which were noted in the previous series, and consisted of enhanced argentophilia and swelling of the synapses. A careful repeat test of this result again confirmed this; no changes of the nerve fibers on the 1st to 4th day, as L. A. Afrikanova [1] reported, were observed.

As distinct from the preceding series, the destructive changes in this case progressed very rapidly, and on the 9th day the majority of the synapses were greatly changed (see figure, b). In this figure a pale zone of neuroplasm is clearly seen around one of the synaptic endings. In comparison with the normal, this pale halo had a less distinct outline.

A fact worth noting is that even after exposure to twice the lethal x-ray dose (1200 r), synaptic endings with little damage occurred along with the disintegrating synapses. This fact probably indicates the unequal radioresistance of the sympathetic neurons of the lateral horns of the spinal cord. The same, or an even greater difference probably exists between the central and peripheral sympathetic neurons, since much greater damage was observed in the preganglionic fibers than the postganglionic fibers after exposure. The microscopic picture in the experiments was very reminiscent of that found in nerve ganglia three to four days after transection of the preganglionic fibers. The difference consisted in the terminal part of the preganglionic fibers after irradiation



Synapses on neurons of superior cervical ganglion of cats at different times after exposure to different doses of x rays: a) argentophilic and swelling of some of the synapses on 7th day after exposure of cats to dose of 500 r. Impregnation by Bielschowsky-Gros-Lawrentjew technique. MBI-3, obj. 100 ×, oc. 10 ×; b) swelling and granular disintegration of synapses, fragmentation of preganglionic fibers 9 days after exposure to dose of 1200 r. Golgi-Deineka treatment. MBI-3, obj. 100 ×, oc. 7 ×; c) two-ring synaptic ending 31 days after exposure to dose of 500 r. Impregnation by Bielschowsky-Gros-Lawrentjew technique. MBI-3, obj. 100 ×, oc. 7×; d) restored synaptic endings 80 days after exposure to dose of 500 r. Golgi-Deineka treatment. MBI-3, obj. 100 ×, oc. 7×.

being altered much more than the distal part. Frequently we found only slightly altered nerve fibers in the sympathetic trunk caudad of the ganglion when the intercellular plexus and synapses were at the stage of disintegration.

Some of the changes which we observed in the synapses after irradiation were obviously reactive changes and, hence, reversible, and some were irreversible. For instance, in cats of the second series there was a considerable reduction in the number of synapses with enhanced argentophilia on the 16th day after exposure. The degree of their argentophilia was also reduced. On the 30th day after exposure none of the observed synapses differed in any way from the controls. Thus, in the animals of this series, there was a gradual restoration of the structure of the synapses in the course of a month.

Besides the gradual decline of the enhanced argentophilia of the altered synaptic endings after exposure of the animals to a dose of 500 r, we more than once managed to observe two-ring synapses, which some authors [9] regard as structures due to progressive-regressive changes. Moreover, from the 30th day after irradiation we discovered growth bulbs of nerve fibers in the sympathetic trunk and among the nerve cells in the ganglion. The figure (c) shows a two-ring synaptic ending found in animals of the third series on the 31st day after irradiation. The pronounced argentophilia of the first ring, and the change in the fibril connecting these rings can be seen. Later, according to Kirsche [8], the first synaptic ring disintegrates completely, and the one following it grows up to the innervated substrate and restores the damaged contact between the neurons.

If, in this case, degeneration affects not only the ending, but also the distal part of the fiber also, fibers grow into the ganglion from the remaining proximal part $1\frac{1}{2}$-2 months after exposure. We observed the growth bulbs of these fibers. Thirty days after exposure the greater number of the revealed synapses were even more modified in shape, size, and color. At this same time, the ganglion revealed many regenerating preganglionic fibers, which had the form of fine threads with frequent small thickenings. The synapses observed 80 days after exposure differed in no way from the normal (see figure, d).

In the fourth series we found no progressive changes in the synapses. The animals in this experiment perished 11-14 days after exposure.

Much still remains obscure in the mechanism of synaptic restoration after the injurious action of x rays, and we will continue our work in this direction.

SUMMARY

The author studied the condition of interneuronic synapses in the superior cervical sympathetic ganglion of adult cats after a single total x-ray irradiation in doses of 30, 100, 500, and 1200 r. Argentophilia of the synapses was noted on the 9th day after the irradiation in the animals (100 r). On irradiation with a dose of 500 r, such changes occurred by the 7th day, whereas, with a dose of 1200 r, on the 5th day. Comparison of the synaptic structures according to the series on the 9th day after irradiation demonstrated that the morphological changes were greater, the higher the irradiation dose. Synapses were restored in 2-2.5 months in the animals which survived, after irradiation with sublethal doses.

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