Threshold and Pattern of Electroshock Seizures after 250 r Whole-Body X-Irradiation in Rats.* (26911)

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Moderate doses of X-irradiation modify the responses of nervous tissue to electrical stimulation(1,2). Exposure to 500 r whole-body X-irradiation in rats lowers the threshold to electroshock convulsions(3-6), alters the pattern of tonic-clonic seizures and delays the recovery of normal brain function after a maximal convulsion(4-6). Furthermore, the changes in convulsive reactivity persist for at least 180 days after a single exposure to 500 r X-irradiation(5,6).

At present these functional changes cannot be explained by morphological alterations because previous work has indicated that doses above 1000 r are necessary in order to affect cellular morphology in the central nervous system(7). It is possible that systemic and local metabolic alterations following X-irradiation could be responsible for these effects on brain function. For example, body growth is retarded after 500 r X-irradiation. However, the reduction in body weight alone can not explain the lower than normal threshold(6). The effects of exposure to low to moderate X-ray doses on brain function have not been clearly defined nor are the underlying mechanisms completely understood in terms of the available data.

The present study was undertaken to compare the effects of 2 doses (250 and 500 r) of X-irradiation on electroshock convulsions for a period of 20 days post-irradiation in rats.

Materials and methods. Animals. Thirty adult male Long-Evans rats of about 240 g body weight at the beginning of the experiment were divided into 3 groups of 8-13 animals each as follows: (1) control, electroshocked; (2) 250 r, electroshocked; (3) 500 r, electroshocked. Food (Purina Fox Chow Pellets) and water were given *ad libitum* except for a 12-hour period of fasting prior to irradiation.

Body weights were recorded periodically for all animals throughout the experiment.

X-irradiation. The experimental animals were exposed to single doses of whole-body irradiation at 13 r/minute with a 250 kv 15 ma X-ray machine (filters: 0.5 mm Cu and 1.0 mm Al).

Electroshock technics. The apparatus for producing electroshock seizures in rats is a constant-current stimulator delivering a sinusoidal 60 CPS current. Woodbury and Davenport(8) have described this stimulator and the various electroshock methods in detail. The following methods are used in this experiment: 1. *minimal electroshock seizure threshold (EST) test,* as a measure of brain excitability; 2. *maximal electroshock seizure (MES) pattern test,* as a measure of spread of excitation; and 3. *return of the righting reflex (RRR) test,* as a measure of brain recovery processes.

The EST is defined as the minimal amount of alternating-current in milliamperes (ma) delivered for 0.2 second through corneal electrodes which will elicit just barely detectable clonic convulsions of ears, jaws, and forepaws without loss of equilibrium and lasting only a few seconds. The MES is produced by delivering 150 ma for 0.2 second through corneal electrodes. With this supramaximal stimulus, normal rats show typical maximal (tonic-clonic) convulsions. Total duration of the seizure, duration of the tonic phase with its flexor and extensor components and duration of the clonus are determined for each animal. The RRR test measures the duration of the depression period

* This paper reports research undertaken in cooperation with the Quartermaster Food and Container Institute for the Armed Forces, QM Research and Engineering Command, U. S. Army, and has been assigned No. 2128 in the series of papers approved for publication. The views or conclusions contained in this report are those of the authors. They are not to be construed as necessarily reflecting the views or indorsement of the Dept. of Defense.

TABLE I. Effect of 250 r and 500 r Whole Body X-Irradiation on Body Weight of Rats.

Group treatment		Body wt, g — Days before irradiation	Days post-irradiation					
		4	0	2	5	10	15	20
Control (13)*	Mean	251	238	265	280	289	298	306
	S.E.	7.2	4.4	5.1	6.2	6.6	7.7	4.8
	% change†		−5	6	11	15	19	22
250 r (8)	Mean	246	229	238 §	243 §	250 ‡	260	287
	S.E.	7.8	7.1	9.2	11.9	18.0	22.4	18.3
	% change		−7	−3	−1	2	6	17
500 r (9)	Mean	246	227	232 §	237 §	222 §	241 §	253 §
	S.E.	5.0	4.9	6.0	7.8	13.9	15.0	13.3
	% change		−8	−6	−4	−10	−2	3

* No. of animals in parentheses.
† % change is computed from pre-irradiation value.
‡ *t* test comparison with controls $P \leq .10$.
§ " " " " $P \leq .05$.

elapsing between the end of the MES and the spontaneous rise of the animal.

In the present experiment the EST was determined 4 times at intervals of 48 hours prior to any experimental manipulation in order to obtain a baseline for all animals. Following fasting and irradiation of groups 2 and 3 and fasting of group 1, EST determinations were made within 5 hours after X-ray exposure, daily for the next 5 days, and then every 48 hours until the end of the experiment on day 20. MES pattern and RRR were assessed on day 5 and 20 post-irradiation.

Results. At 20 days post-irradiation the mortality for the 250 r and 500 r irradiated groups was 1.4% and 2.2%, respectively.

The pattern of body growth indicated by changes in body weight is presented in Table I. Control rats grew at a normal rate characteristic for the age, sex and strain of the animals; it has already been shown that electroshock alone does not alter body growth (6). Irradiation at 500 r significantly (by the *t* test) retarded the rate of body growth as compared to controls from day 2 to 20 post-irradiation. The difference between body weights of controls and of the 250 r group was statistically significant from day 2 to 10 post-irradiation, *i.e.* for a shorter period of time than after 500 r.

Control rats demonstrated the progressive increase in threshold after the initial fall due to fasting shown to be associated with increasing age(9). After 500 r and 250 r irradiation this normal increase was inhibited, and irradiated rats maintained a lower threshold than non-irradiated controls for the duration of the post-irradiation period (Fig. 1). The difference between irradiated and control rats became statistically significant at about day 8 post-irradiation and remained significant to day 20. There was no difference in threshold between the 2 levels of irradiation.

Irradiation at doses of 250 and 500 r had no significant effect on maximal seizure pat-

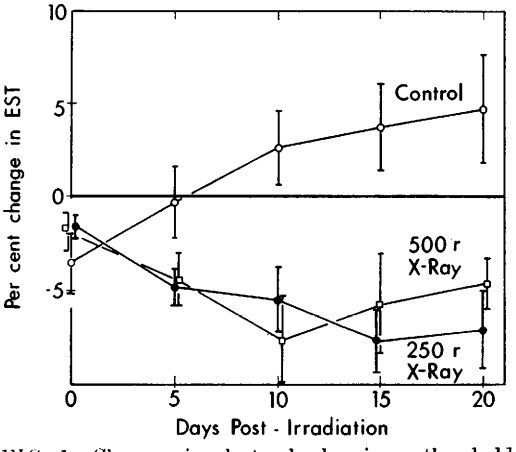

FIG. 1. Changes in electroshock seizure threshold (EST) for a 20-day period for control, 250 r, and 500 r groups. Data are presented as percent of change from a pretreatment value taken at one day before irradiation and/or fasting. Stand. errors are indicated by bracketed lines.

TABLE II. Effect of 250 and 500 r Whole-Body X-Irradiation on Maximal Seizure Pattern of Rats 20 Days after Exposure.

Group and treatment		Maximal seizure pattern (duration of phases in sec.) Tonus	Clonus	Total seizure
Control (12)*	Mean	14.4	32.0	46.5
	S.E.	.35	3.5	3.5
250 r (7)	Mean	13.4	18.9‡	32.3‡
	S.E.	.42	1.8	1.5
500 r (7)	Mean	14.5	23.7†	38.2†
	S.E.	.53	3.2	2.7

* No. of animals in parentheses.
† *t* test comparison with controls indicated P ≤.10.
‡ *t* test comparison with controls indicated P ≤.05.

tern on the 5th day post-irradiation. By day 20 post-irradiation the duration of the total maximal seizures was reduced and this was the result of a shorter clonic phase (Table II). In the present experiment postictal depression was not modified to a statistically significant degree by irradiation. However, the irradiated rats tended to have a longer period of depression after maximal seizures than control animals. This finding has been described previously(6).

Discussion. Single exposures of rats to whole-body X-irradiation in moderate doses can alter the threshold for electrically induced seizures. With the methods used in these experiments, increased convulsability is indicated by a lower than normal threshold. The data presented show that both 500 r and 250 r irradiation inhibit to the same degree the characteristic progressive increase in threshold with age and produce similar changes in the maximal seizure pattern.

It is interesting that there have been a number of studies indicating a plateau in response to a range of radiation doses. Quastler *et al.*(10) showed that a 3½ day mortality for mice was a stable finding in a dose range of 500 to over 4000 r. In a study of abdominal sensitivity to X-ray in rats (Bond *et al.* 11), a plateau was observed in average day of death and in body growth over a dose range of 1600 to 2200 r. Quastler and Baer (12) working with irradiated seeds of Mung beans showed a degree of inhibition of growth which was stable over various dose ranges. Finally in the study of brain electrical activity by Gangloff and Haley mentioned above (1), the same magnitude of change was observed after 200 and after 400 r.

The works cited together with the results of the present experiment suggest that irradiation-induced changes in several systems occur to a maximum extent after a threshold dose of radiation is exceeded. These changes may involve a particular mechanism which manifests an all-or-none response to radiation. Increasing the radiation dose above the threshold level would have no added effect unless or until a second mechanism is affected by the radiation.

In view of the retardation in body growth after irradiation it might be possible to correlate general metabolic alterations with functional changes in the central nervous system. However, evidence has been presented that there is no direct relation between alterations in rate of body growth and changes in threshold after irradiation(6). Similarly in the present experiment (Table I and Fig. 1), the fact that body weight was reduced to a greater extent by the higher radiation dose whereas changes in brain function were of the same magnitude with both doses supports the hypothesis that these phenomena are somewhat independent expressions of selective systemic and local metabolic processes.

The increased excitability after irradiation indicated by the lower than normal thresholds is also apparent from some of the aspects of the maximal seizure pattern (Table II). For example, in all irradiated groups the ratio between lengths of tonus and clonus was greater than in the controls. On the other hand, the shorter total duration of the maximal seizures in the experimental animals as compared to controls suggests interference by irradiation with the spread of nervous impulses in the central nervous system. Shortening of maximal seizures, together with prolongation of the postictal depression could be interpreted as an increase of central inhibition or as a greater than normal degree of fatigue of the neural elements after heightened activity. This interpretation which has been

proposed in relation to the effects of 500 r (6) seems to be valid for the smaller dose of 250 r.

Summary. The effects of single doses of 250 and 500 r whole-body X-irradiation on threshold and pattern of electroshock seizures were followed in rats for 20 post-irradiation days. Irradiated rats had lower electroshock seizure thresholds than controls. Total duration of the maximal electroshock seizures was shorter in the irradiated rats than in controls. This was due to a significant shortening of the clonic phase of the seizure. These findings are interpreted as indicating that brain excitability is increased by irradiation at 250 and 500 r. However, heightened seizure activity cannot be sustained as long in irradiated rats as in controls. The magnitude of the changes in brain convulsability was the same for the 2 doses of X-irradiation while the effects on body growth were dose-dependent in this range.

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Received July 28, 1961. P.S.E.B.M., 1961, v108.