# CHANGE IN CHOLINESTERASE ACTIVITY IN ACUTE RADIATION SICKNESS

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The view that radiation sickness results from a failure of coordination of the different organs and systems makes it necessary to examine, carefully, disturbances of the basic regulatory mechanisms occurring in this disease. First in importance are the enzyme systems, and in particular cholinesterase, the enzyme responsible for mediating central nervous excitation and its transmission to effector organs.

In the present work, an attempt has been made to study cholinesterase activity in several different tissues.

## METHOD

The experiments were carried out on mice and on frogs. The mice were exposed to gamma radiation from radioactive mesothorium, the dose being 700 or 10 000 r ± 10%. When irradiated with 700 r, all the mice died within a month. After irradiating with 10,000 r, all the animals died within 3-4 days. Cholinesterase activity measurements were made in the tissues of the brain, liver, and intestine, and in addition, the sensitivity of the irradiated mice to strychnine was measured.

In experiments on frogs, an ampoule containing 100 mC units of mesothorium (lead filter 1 mm) was sewn in position over a vertebra. The irradiation was continued for 3-5 days. On the 14-16 day, an abscess had formed at the site of the ampoule. The animals died after 2-3 weeks. Convulsions occurred one week after the irradiation. In frogs, measurements were made of the cholinesterase activity in the spinal cord, and changes in the sensitivity to strychnine and neostigmine were noted. Strychnine was given intraperitoneally to mice as a 1: 50,000 and to frogs as a 1: 10,000 solution, the dose being the same per g body weight. Neostigmine diluted 1: 1000 was injected intraperitoneally into frogs.

Cholinesterase activity was measured using R. Ammon's method [2] and a Warburg's apparatus.

## RESULTS

Measurements of cholinesterase activity. Table 1 shows results of measurements on the spinal cord of irradiated and control frogs.

From Table 1 it can be seen that at the end of the irradiation, there is an immediate drop in cholinesterase activity of 35% as compared with the controls. It continues to fall, and after 2 days has reached 40% of the control value, i.e., it has been reduced by 60%.

Table 2 shows the results of corresponding measurements in control mice.

It can be seen that the greatest activity is found in the liver, and the least in the brain. The activity is more constant in the latter.

Table 3 shows the results of measurements of cholinesterase activity in mouse tissue after irradiation with 10,000 r. These results must be considered as appoximate, on account of the small number of experiments and the comparatively wide scatter in the results obtained. However, a comparison is shown between these results and those of Table 2, and it can be seen that after irradiation, there is a reduction in cholinesterase

TABLE 1

Cholinesterase Activity in γ of Hydrolyzed Acetylcholine per 100 mg Spinal Cord per 40 Minutes

| No. of animal | Control | Immediately after irrad. | Two days after irradiation |
|---|---|---|---|
| 1 | 2 057 | 2 43 | 1 050 |
| 2 | 1 940 | 2 360 | 1 740 |
| 3 | 2 200 | 1 770 | 1 700 |
| 4 | 2 070 | 2 170 | 1 050 |
| 5 | 2 070 | 1 323 | 1 220 |
| 6 | 1 740 | 1 270 | 540 |
| 7 | 2 800 | 1 280 | 1 100 |
| 8 | 2 600 | 1 803 | 870 |
| 9 | 2 600 | — | — |
| 10 | 2 700 | — | — |
| Average | 2 277 | 1 482 | 927 |

TABLE 2

Cholinesterase Activity in γ of Hydrolyzed Acetylcholine per 100 mg Tissue per 40 Minutes

| No. of animal | Liver | Brain | Intestine |
|---|---|---|---|
| 1 | 2 426 | 512 | 1 070 |
| 2 | 1 552 | 627 | 1 326 |
| 3 | 2 820 | 672 | 1 443 |
| 4 | 2 692 | 588 | 1 364 |
| 5 | 2 840 | 617 | 723 |
| 6 | 3 070 | 672 | 987 |
| 7 | 2 046 | 578 | 1 435 |
| 8 | 2 112 | 526 | 1 048 |
| 9 | 2 334 | 515 | 946 |
| 10 | 2 198 | 613 | 947 |
| Average | 2 410 | 592 | 1 129 |

TABLE 3

Cholinesterase Activity, in γ of Hydrolyzed Acetylcholine per 100 mg of Tissue per 40 Minutes

| No. of experiment | Liver | | | | Brain | | | | Intestine | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Time (in days) after irradiation | | | | Time (in days) after irradiation | | | | Time (in days) after irradiation | | | |
| | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 |
| 1 | 1 260 | 2 088 | — | — | 752 | 1 265 | — | — | 936 | 1 315 | — | — |
| 2 | 1 476 | 1 882 | 2 838 | 2 532 | 391 | 534 | 312 | 259 | 1 132 | 923 | 1 491 | 880 |
| 3 | 1 468 | 2 266 | 2 038 | 1 194 | 245 | 347 | 253 | 300 | 1 157 | — | 1 034 | 775 |
| Average | 1 403 | 2 078 | 2 438 | 1 863 | 492 | 715 | 282 | 279 | 1 075 | 1 119 | 1 262 | 817 |

activity. For the liver, this reduction was greatest on the first and fourth days after irradiation, being 30 % on the first day and 20% on the fourth, while for the brain the corresponding figures were 17 % and 50%; for the intestine, there was a 25% reduction by the fourth day.

Table 4 shows the results of cholinesterase activity measurements in mice irradiated with 700 r.

These results show that this dose causes an appreciable change. The reduction is on average:

| Organ | First Week | Second Week | Third Week |
|---|---|---|---|
| Liver | 22 % | | 23 % |
| Brain | 28 % | 14 % | |
| Intestine | 19 % | | 24 % |

The effect of strychnine and neostigmine on convulsions in irradiated animals. Frogs in which the cord was irradiated showed an increased sensitivity to strychnine and to neostigmine. In the control group of 50 animals, convulsions occurred 20 minutes after the injection of 2γ per g of strychnine, or 10γ per g of neostigmine.

The spinal cord was irradiated in 40 frogs for 3 days; 1 per g of strychnine was injected into 20 of these and 5γ per g into the remaining 20, and in each case convulsion occurred after 20 minutes.

TABLE 4

Cholinesterase Activity, in γ of Hydrolyzed Acetylcholine per 100 mg of Tissue per 40 Minutes

| No. of experiment | Liver | | | Brain | | | Intestine | | |
|---|---|---|---|---|---|---|---|---|---|
| | Time (in weeks) after irradiation | | | Time (in weeks) after irradiation | | | Time (in weeks) after irradiation | | |
| | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 |
| 1 | 1 226 | 3 102 | 1 470 | 430 | — | 464 | 808 | 1 106 | 845 |
| 2 | 1 464 | 2 304 | 1 836 | 304 | 540 | 595 | 923 | 1 198 | 805 |
| 3 | 2 862 | 2 580 | 2 186 | 441 | 390 | 403 | 945 | 1 026 | 607 |
| 4 | 2 000 | 2 204 | 2 052 | 461 | 540 | 691 | 784 | 1 201 | 1 132 |
| 5 | — | 2 008 | 1 770 | 523 | 631 | 476 | 1 176 | 1 022 | 918 |
| Average | 1 880 | 2 430 | 1 860 | 432 | 520 | 525 | 922 | 1 110 | 861 |

No increased sensitivity to strychnine was found to follow irradiation in mice. In the control group, the convulsions occurred 20 minutes after injecting 0.08 ml of a 1: 50,000 solution per g weight of strychnine (10 animals); 0.05 and 0.07 ml of the same solution caused only an increased excitability over the same period (20 animals used). After irradiating with 700 r, an injection of 0.05 ml of strychnine per g again caused only a small increase in excitability for the first 4 weeks after irradiation (50 animals). The same result was observed during the first 3 days after irradiating 30 frogs with 10,000 r. It was interesting that right up to the time of death, there was no increased sensitivity to strychnine.

From our experiments it follows that for both frogs and mice, the activity of cholinesterase is reduced after irradiation. The greatest fall in activity in mice corresponds with the period of the primary reaction and the time of manifestation of acute radiation sickness. The great reduction in the cholinesterase activity in frogs as compared with mice may be due to the much greater dose of radiation. There is no doubt that cholinesterase activity in tissues is reduced after irradiation, and confirmation has been given in other published reports.

Burn, Kordik, and Mole [3] found a reduced pseudocholinesterase activity, and Conard [4] found a reduced cholinesterase activity in the intestine of a rat receiving a total radiation dose of 500-1000 r. Nachmansohn [5] attributes changes in the acetylcholine — cholinesterase system to the effect of the ionizing radiation on the nervous conducting elements.

The experiments on the development of convulsions in mice following strychnine injections, showed that irradiation did not raise the sensitivity to this drug. However, there was an increased sensitivity to strychnine in the same experiment in frogs. Possibly, the difference may depend on the dose, and on the region irradiated, which, in frogs, was the spinal cord. Also, in frogs, radiation by itself may induce spasms, and strychnine merely facilitates their development, while in warm-blooded animals not many authors have observed definite convulsions, and these occurred only just before death from very large doses.

## SUMMARY

Total gamma irradiation of mice with a dose of 700-1000 r provoked a decrease of cholinesterase activity in brain, liver and intestine; this occurred during the first and third week after irradiation, during the period of the primary reaction when the disease was at its height; the change was especially pronounced shortly before death.

Total gamma irradiation with 700-10,000 r causes an increased sensitivity to strychnine in frogs, but not in mice.

## LITERATURE CITED

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