Responses of Swine to High Doses of Radiation. (24837)

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Small laboratory animals have been studied extensively at high radiation doses(1,2) and a few results with monkeys(1), dogs(3) and swine(4) have been reported. Because of some striking differences in response patterns of different species more data are desirable. Large animals having cross sections comparable to that of man are of particular interest since similar depth dose distributions can be obtained. Laboratory studies at high doses have been limited by the requirement for high dose rate over a large field, so that exposure times will be short compared to survival times. With high intensity X-ray source available, high dose studies at reasonable rates became possible in mammals as large as swine, which have been utilized in this study. In addition to adding detailed information on another species, use of swine provided better data on high-dose incapacitation than is obtainable from smaller animals.

Methods. Twenty-six cross-bred swine 3- to 4-months-old, weighing from 80 to 130 lb. were used. Pre-irradiation hemograms and complete physical examinations were done on all swine, and all were in good health at time of irradiation. The X-ray source and calibration procedures have been described(2). Swine irradiations were done at 3.0 to 3.5 Mev constant potential beam having an HVL of 21 cm in masonite phantom. With target-skin distance of 180 cm, dose rate in air at mid-swine position was 150 r/min. Special filters gave a radiation field flat to ± 5% to a radius of 2½ ft. which permitted simultaneous exposure to 4 swine. At highest doses, exposure times were an appreciable fraction of survival times calculated from the mid-point of irradiation. Behavior during irradiation was observed with a closed-circuit television system. After irradiation all animals were under constant observation until death. Blood samples were collected from anterior vena cavae for hematological studies by standard methods. Properdin titers of sera of some swine were determined.* Sections of stomach and intestines were removed immediately after death and complete autopsies performed within 2 hours. Tissues were examined grossly and microscopically.

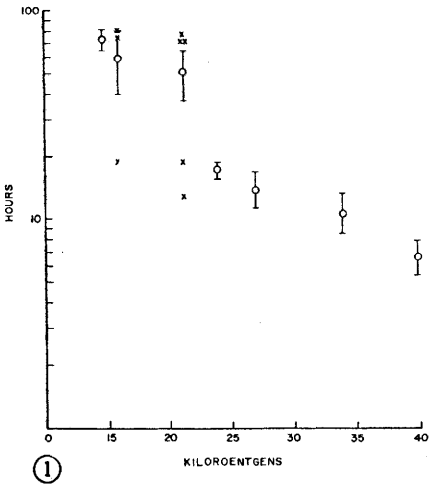
Results. At this dose rate the first sign of radiation injury was restlessness, starting at 2000 to 2500 r and increasing with dose. At 3500 to 4000 r (22 to 27 minutes) acute distress was evidenced by extreme salivation, mastication, retching and profuse emesis. Diarrhea appeared at about 15,000 r. With exception of diarrhea, these manifestations ceased at 10,000 to 20,000 r and were followed by calm which progressed to a marked depression.

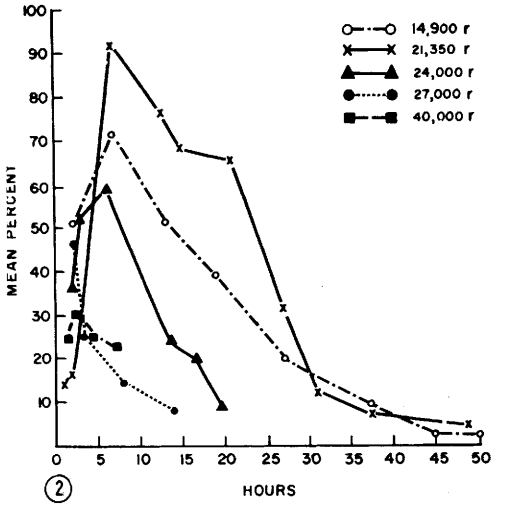
Although depressed, the swine recognized auditory and visual stimuli after 20,000 r (133 minutes) and were capable of responding to them. Depression continued to increase with dose, however, and by 30,000 r (200 minutes) all swine were prostrate. They did not respond to auditory, visual or dermal stimuli. The conjunctival reflex was absent and ataxia was evident when the swine attempted to rise. Transient central nervous system manifestations appeared at 39,000 r (260 minutes) with convulsions and loss of equilibrium.

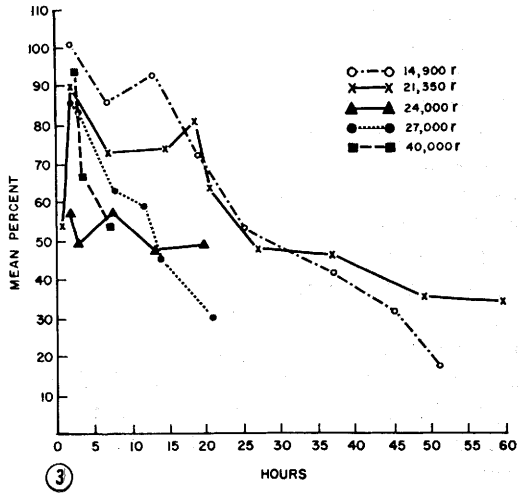
Swine normally have a rectal temperature of 101.6 to 103.6°F, pulse 60 to 80/min and respiratory rate of 8 to 18/min. By 14,900 r mean values had reached 105.0 ± 0.2°F, 169 ± 26/min and 67 ± 17/min. At this stage respirations were irregular with prolonged expiratory phase. Respiration became increasingly rapid and shallow as dose progressed.

At doses of 21,350 r and below, the swine were markedly depressed and moderately ataxic when exposure was terminated. Some presented diarrhea and vomiting. Of interest was their striking improvement from 1 to

* Courtesy of Dept. of Serology, Walter Reed Army Inst. of Research.







3 hours after exposure. Even though diarrhea and vomiting persisted, several ate, drank small amounts and foraged through the bedding. It appeared that at least simple, and probably more complicated, tasks could have been performed. This period of "well-being" persisted 10 to 48 hours in various animals, after which their condition deteriorated. Thereafter, death ensued within a few to 24 hours. The 24,000 and 27,000 r groups presented minimal recovery and were more like higher dose groups in their response.

The highest dose swine (40,000 r) were completely incapacitated after exposure. Mean rectal temperature was 106.6 ± 0.3°F, pulse rate 172 ± 11/min, and respiration 84 ± 23/min. They were prostrate and even when assisted could not stand. Nystagmus developed within an hour or 2 and pupillary response to light was considerably diminished. Transient seizures appeared from 10 minutes to 3 hours after completion of exposure. These progressed from transient episodes of clonic spasms of various muscle groups and false "running" movements to severe convulsions and opisthotonos. Pulse rate remained approximately twice normal, respiratory rate 4 to 5 times normal and temperature response varied throughout the group. The swine remained completely incapacitated. Death occurred during violent convulsions. The 34,000 r group was similar in response.

Fig. 1 presents mean survival time by dose group. Standard errors are indicated. The wide limits at 16,000 r and at 21,350 r result from a definite "split" in survival time, *i.e.*, these swine either died in less than 20 hours or survived more than 70 hours. Individual survival times at these doses are given in Fig. 1.

Mean leukocyte responses are presented in Fig. 2. For clarity in the Figure, responses

FIG. 1. Survival times of swine exposed to various supralethal doses of X-radiation. Mean and stand. error of mean are indicated for each dose. Individual points plotted for 2 doses due to definite "split" in survival times.

FIG. 2. Circulating leukocyte response of supralethally irradiated swine at various times after exposure. Plotted points are mean percents of mean pre-irradiation values.

FIG. 3. Blood platelet response in swine at various times after supralethal irradiation. Points are mean percents of pre-irradiation mean values.

at 16,000 r and at 34,000 r are omitted since they closely resembled those at 14,900 r and 27,000 r respectively. In all, swine lymphocytes were drastically reduced on first post-irradiation sample and were at minimal levels by the 12-hour sampling. The relative proportion of segmented neutrophils was increased immediately after irradiation as was the absolute count. This rapidly changed to a predominance of band forms. Myelocytes and metamyelocytes were present immediately after irradiation and persisted about 36 hours or until death. In many swine normoblasts were present immediately post-irradiation. Though decreased in numbers these persisted for as long as 12 hours. After about 36 hours, eosinophils became the predominant cell type. Many of these were eosinophilic metamyelocytes. In some swine toxic granulation of neutrophils appeared as early as 26 hours. Those swine surviving more than 70 hours presented a profusion of basket cells prior to death.

Mean blood platelet responses are presented in Fig. 3. As in Fig. 2 16,000 r and 34,000 r groups are omitted.

Erythrocyte counts of only the 14,900 r group decreased at 36 hours post-irradiation (16,000 r unsatisfactory). Mean corpuscular volume, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration values were not significantly different after irradiation. Before irradiation erythrocyte fragility values were 0.608 $\pm$ 0.0058 and 0.433 $\pm$ 0.0042 for partial and complete hemolysis respectively. At all times after irradiation the 34,000 r swine (40,000 r not done) presented significantly increased fragility. Mean value for partial hemolysis was at least 0.690 (endpoint missed in some samples) and was 0.493 $\pm$ 0.0053 for complete hemolysis. By 8 hours after 27,000 r respective mean values for partial and complete hemolysis were at least 0.651 (end-point missed) and 0.461 $\pm$ 0.0126. This increased fragility persisted until death. Other dose groups did not present significantly altered erythrocyte fragility. In view of exposure time reticulocyte (proerythrocyte) values are considered for only the 3 lower dose groups. Based on mid-point of sampling interval, these disappeared at 24 hours in the 21,350 r and the 16,000 r groups and were absent by 34 hours in the 14,900 r group.

Hematocrit levels decreased after irradiation although the changes were not statistically significant. In most cases terminal samples presented hematocrit values much higher than normal. The uncorrected erythrocyte sedimentation rate increased drastically and continuously from 24 hours after irradiation until death. Not infrequently these values were in the range of 45 to 55 mm.

The properdin response in blood serum in the 14,900 r, 24,000 r, 27,000 r and 34,000 r groups was characterized by mean decrease of 32% from 12 hours onward. If all samples on all swine for 12 hours and more after irradiation are combined, a mean of 4.8 units/ml, as compared with pre-irradiation mean of 7.1 units/ml, results. This difference is statistically highly significant ($p \leqq 0.005$).

Discussion. Although the numbers of swine are not large, these data suggest an abrupt survival time transition as in the guinea pig or mouse, rather than the smooth semi-log response of rat and hamster. This is borne out by the shift from depression to convulsions and to some extent by blood data. The abrupt transition in mean survival time between the 21,350 r and 24,000 r groups with an exponential relationship at doses above the transition zone undoubtedly reflects a different mode of death. Although occurring at a point intermediate between those for guinea pig (6 Kr) and mouse (43 Kr), it is unlikely that the phenomenon is different.

The lower dose rate can probably be considered as responsible for only slight evidence of Langham *et al.*'s acute ataxic phase(1). Their lethargic, hyperactive and terminal phases were in evidence. Even more similar was the clinical response of guinea pigs(5,6). At doses above 30,000 r, swine remained completely incapacitated throughout the brief post-irradiation period. It appeared that the minimal recovery at 24,000 r and 27,000 r would not have allowed performance of other than the most basic tasks, if any. Following a period of 1 to 3 hours after irradiation, swine receiving lower doses presented dramatic recovery and undoubtedly could have func-

tioned with only partial impairment until 10 to 48 hours after irradiation. During exposure, accumulation of about 4,000 r greatly impaired function as a result of nausea and severe emesis.

These and previously cited data emphasize the differences in species and leave the response of man unknown. It may be of significance that in Japan there was no mention of an excitatory phase but depression was of common occurrence. Practically, it may make little difference, for radiation depressed humans may be able to function for varying periods because of cerebral motivation questionably present in animals. Further, man may be reasonably functional between convulsions. Thus, evaluation of incapacitation in these swine may not be indicative of that occurring in heavily irradiated human population.

At this dose rate granulocytosis was not observed immediately post-irradiation. Transient leukocyte recovery at lower doses was undoubtedly due to release of immature forms into the circulating blood. The presence of numerous basket cells by the third day suggests that destruction of the mechanism for removal of damaged neutrophils may occur at doses lower than previously reported(1).

Platelet counts start to decline about the third or fourth day after mid-lethal range irradiation. Since decreases were initiated much earlier in these swine, damage to platelet forming and release mechanisms must have been more profound. These responses suggest dose dependency.

Increased fragility of erythrocytes in the 27,000 and the 34,000 r swine demonstrates a decreased resistance to hemolysis after massive doses of irradiation. Osmotic fragility of young mature human erythrocytes is less than that of older cells(7). Even if the same is true for swine, it is difficult to conclude that the cell population studied was so largely composed of older erythrocytes since increased fragility was present soon after irradiation. It seems unlikely that increased fragility can be attributed to a greater radiation susceptibility of younger erythrocytes; thus, the mechanism remains in doubt. The slight decrease in hematocrit may be indicative of erythrocyte destruction. The terminal increase in hematocrit was undoubtedly due to hemoconcentration.

Significance of the properdin system in the response to mid-lethal irradiation remains in doubt(8,9). In swine reported herein, a decrease occurred earlier than for mid-lethal irradiation in other species, but did not continue. Relation to survival time could not be demonstrated. The significance of the decline is difficult to assess.

The gross and microscopic pathological changes will be reported elsewhere.

Summary. Although time and dose relationships were different, the response of swine to high doses of radiation was similar to that of guinea pigs and mice. For practical purposes, at doses of 24,000 r and above, incapacitation was complete in these swine. It is conceivable that if these animals possessed cerebral motivation similar to that in man, capabilities might have been greater. At lower doses a striking recovery occurred within 1 to 3 hours after exposure.

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