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# Effects of Suspended Animation on the X-Irradiation Syndrome<sup>1</sup>

#### JAMES P. ROBBIE<sup>2</sup>

Abstract. Male albino mice in a condition of suspended animation were X-irradiated with a dosage of 2,000 r. These animals showed a remarkably greater resistance to the X-rays than did controls that were X-irradiated only. The animals that had been in suspended animation often lived twice as long as the controls. However, in many cases they died from shock shortly after being revived.

Recently it has been found that rats and mice can be cooled until their internal temperature approaches  $0^{\circ}$ C and still be successfully revived. Until Andjus (1951) accomplished this feat, it was believed that if the internal temperature of an adult non-hibernating mammal dropped below about 15° C, the animal would die from hypothermia. When a rat's body temperature drops below 15°C or a mouse's below 5°C, respiration and circulation both cease, and the metabolic rate is considerably slowed down or completely stopped. This condition is called suspended animation. The process of reviving an animal from suspended animation is called reanimation.

Since Andjus' discovery in 1951, he and several others (Andjus 1955; Andjus and Smith, 1955; Goldzveig and Smith, 1956) have been working on methods of improving the survival rate of animals put into suspended animation. Other experimenters, like Smith (1956), have been working with hamsters, and have been reviving them from temperatures below  $0^{\circ}$ C.

Metabolism in an animal under suspended animation is nearly nonexistent. No consistent relationship has been found between metabolism and the X-irradiation syndrome. In 1941 it was found that if the temperature around newborn mice and rats was lowered during X-irradiation of the whole body or of the skin, these animals would be more resistant to the X-rays (Evans *et al.*, 1941). However, Hempelmann *et al.* (1949) found no increase in survival rate by chilling adult mammals.

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The purpose of the study reported here was to determine whether an adult non-hibernating mammal in suspended animation could tolerate X-irradiation better than one given irradiation only.

#### MATERIALS AND METHODS

Sixty male albino mice were put into a condition of suspended animation, then X-irradiated. Each mouse was first put into an airtight pint jar in a "cold room" of about  $5^{\circ}$ C. He was left there until he was in a state of narcosis, which usually took about one hour and twenty-five minutes. This step decreases the body's need for oxygen more effectively than does a normal anesthetic. It also gradually decreases body temperature, instead of abruptly changing it from room temperature to that of ice. The mouse was then taken out of the jar and dipped into ice water. This removed the insulating effect of his fur. Then he was packed in ice, where he was left for one hour. Everything but his nose was buried, since he continued to breathe, at first. After about five minutes, respiration and circulation both ceased.

After he had been on ice for one hour, he was transferred to a cardboard dish containing more ice and moved under the X-ray machine, where he was given 2,000 r of whole body X-irradiation. Irradiation of all mice was done with a General Electric Maxitron X-ray machine operating at 250 kvp. and 30 ma. A .25 mm. copper—1 mm. aluminum filter was used.

To begin reanimation, the mouse was dried off and put under a sixty-watt lamp, where the temperature around him was kept at  $38^{\circ}$ C. He was given mouth-to-mouth artificial respiration through a plastic tube until he started breathing on his own. His heart started beating in about five minutes, and he started breathing about ten minutes later. Then he was put under a lamp where the temperature was kept at  $32^{\circ}$ C.

In addition to the sixty mice that were treated by the above procedure, two groups of controls were used. The first set, consisting of forty-five mice, was anesthetized and then X-irradiated along with the frozen mice. They were used to determine the effect of X-rays on normal mice. The other set of controls, consisting of twenty-two mice, was frozen, then reanimated, without being Xirradiated. This group was used to determine the approximate percentage of mice that died from freezing, not X-irradiation.

#### EXPERIMENTAL RESULTS

It can be noted (Table 1) that five of the mice that were frozen and X-irradiated outlived all of the ones that were X-irradiated only. The bulk of all mice that were not in suspended animation 1960]

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#### Table 1

Incidence of Death of Mice That Were: 1) Frozen Only, 2) X-rayed Only, and 3) Frozen and X-rayed

Groups	Time in hrs.						1	avs			
	No.	0-1	1-24	1-2	2-3	3-4	4-5	5-6	6-7	7-8	8 & over
Frozen only	22	12	3	1	—	1					5
X-rayed only	45		_			32	9	3	1		
Frozen and X-rayed	60	28	19	2	1	3	2		1	4	

and received a dosage of 2,000 r died between the third and the fifth days. The four animals that lived nearly eight days outlived by a factor of two the vast majority of those that were X-irradiated only.

In many instances, however, the mice that had been frozen and irradiated died before the "irradiated only" group. In comparing the instances of death of the frozen and X-irradiated mice with the "frozen only" group it can be noted that X-irradiating the mice does not make it any harder to reanimate them. The percentage that died during the first hour after reanimation was approximately the same in both groups (12 out of 22 or 54.6% in the "frozen only" group and 28 out of 60 or 46.7% in the frozen and X-rayed group). However, the X-irradiation does seem to add to the shock caused by freezing, which is the main cause of death after reanimation. Consequently, only three out of twenty-two or 13.6% died within the first twenty-four hours in the "frozen only" group, whereas nineteen out of sixty or 31.7% died from the combined shock of freezing and X-irradiation. Apparently the shock lingered on into the next few days in a few cases. This accounts for the scattered deaths in the first to the fifth day. Two mice that died on the third day probably would have lived significantly longer, except that they became involved in a fight and killed each other.

#### DISCUSSION

Although not too many mice in the frozen and X-irradiated group outlived the ones in the "X-irradiated only" group, the results are still fairly significant because of the length of time the animals lived beyond the controls, and because the early deaths were caused from the shock of the freezing rather than from the X-rays.

The biggest problem is still to master the technique of successful reanimation. Much work needs to be done to improve the long term survival rate of animals after suspended animation. Andjus, Smith, Goldzveig, and others are working on methods of improving these techniques. When the proper conditions are worked out it should be

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possible to avoid the complications in the results from animals dying from shock rather than from X-irradiation.

To the author's knowledge this is the first attempt of studying the relationship between suspended animation and the X-irradiation syndrome.

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