Survival of Mice During Chronic Centrifugation

I. Studies of Male Mice at Different Ages At Onset of Exposure to One Field and Those At Different Intensities of Gravity for Animals of the Same Age

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GRAVITY has been the most persistent and constant environmental factor throughout the evolution and development of terrestial life forms. Nonetheless, there has been a sparsity of investigations pertaining to the role that this factor plays in the determination of life-span. Until the enthusiasm for space flight was stimulated in 1957, there was little interest in the biological effects of prolonged centrifugation. Most studies have been confined to fields so intense as to provoke death within a period of minutes.[2,4,5]

Increased gravity would impose an added work load upon an animal. This should not only increase his food requirements, but at the same time render the obtaining of food a more arduous task. Internal displacement of body constituents should place the animal at a disadvantage in maintaining a normal life-span. As the animal, through evolution, is adapted to one specific gravitational field intensity, one subjected to this new environmental stress would be expected to live a shorter than normal life. The stress which normal gravity imposes upon even our day-to-day life is well summarized in the words of D'Arcy Thompson:

"In a handful of matter cohesion, capillarity, chemical affinity, electric charge are all potent: across the solar system gravitation rules supreme—Gravity not only controls the actions but also influences the forms of all save the least of organisms. The tree under its burden of leaves or fruit has changed its every curve and outline since its boughs were bare, and a mantle of snow will alter its configuration again. Sagging wrinkles, hanging breasts and many another signs of age are part of gravitation's slow relentless handiwork."[10]

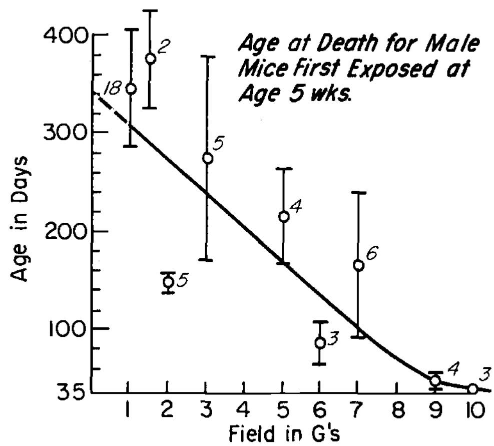

Fig. 1. Numbers beside experimental points indicate the number of animals at each experimental condition. Values at one G are considered to be that of the control. Standard errors are indicated.

Developmental studies with altered G-fields date back to the work of Knight in 1806. He demonstrated that centrifugal fields could orient the direction of growth with bean plants.[7] It was not until 1953 that Stephen Gray pursued quantitative studies with such fields.[6] He found that intense fields decreased the growth rate of wheat coleoptiles. Fifty per cent of the plants survived four days of exposure to 25 G. The first work with animals appeared in 1953. This consisted of an abstract by Matthews[8] stating that although rats could live quite well at 3 G

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they could not attain normal size. Unfortunately, this work has never been described in more detail.

Our laboratory has reported similar results with fly larvae, mice and hamsters.[1,11,12,13] Smith and his co-workers at Davis also have obtained comparable results with chickens.[9] All forms studied demonstrated developmental changes of the type expected to enhance the ability of the animal to survive at high gravity.

With warm-blooded forms, the changes in body mass are of a type one would anticipate upon exposure to any moderately detrimental agent.[1] During the first few days of centrifugation, the animal loses mass. Then the surviving individuals regain part or all of the lost mass, with a growth rate which approaches that of the control rate. Although many individuals grow beyond the initial size, the average experimental animal eventually ceases growth before the control does.

The only published survival data for extended periods of exposure of animals have been those of Smith, Winget, and Kelly working with chickens.[9] After 24 weeks of centrifugation at 2.5 G, 64 per cent of the birds survived. In our laboratory, fruit fly larvae showed almost complete survival during two or three days of growth at 3,000 G. Britton's group report that rats held in a supine position at fields of from 5 to 25 G demonstrate 50 per cent survival after exposure to a constant product of time and field equal to 50 G-minutes.[2] Cranmore[5] reported that at lower fields there exists a so-called "infinite" survival time. Chung[4] reported similar results with mice for exposure as long as 80

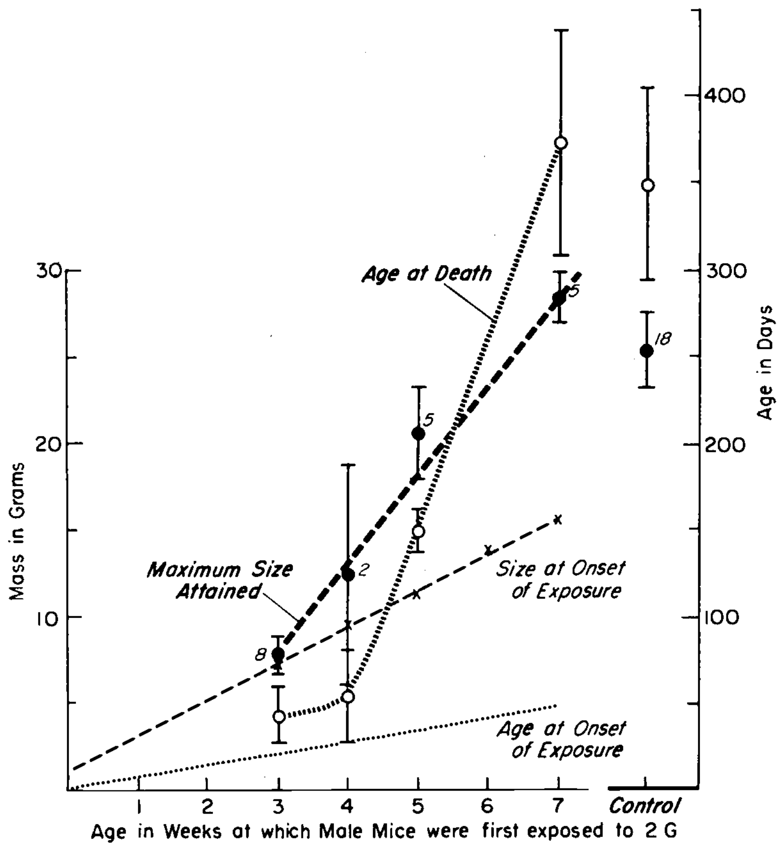

Fig. 2. Influence of age upon ability to tolerate high gravity.

minutes. The purpose of our study has been to investigate survival at fields which permit mice to live for a major portion of their normal life-span.

Chow" for mice and uncooked potatoes as a source of water. After adjusting to the new environment, most mice were able to consume food and resume growth. Mice were not observed during the actual period of centrifugation. Motion pictures of hamsters during centrifugation at 4 G showed an essentially normal behavior. They were seen eating and carrying out other physiological functions. There was only one abnormality in their appearance. Due to the greater field, their movements were more clumsy and ponderous, resembling that of a more massive animal. It can be assumed that mice behave in a similar manner. It is known that some mice adjusted sufficiently to be able to conceive, carry, deliver, and rear litters during their life in the centrifuge.[1,11]

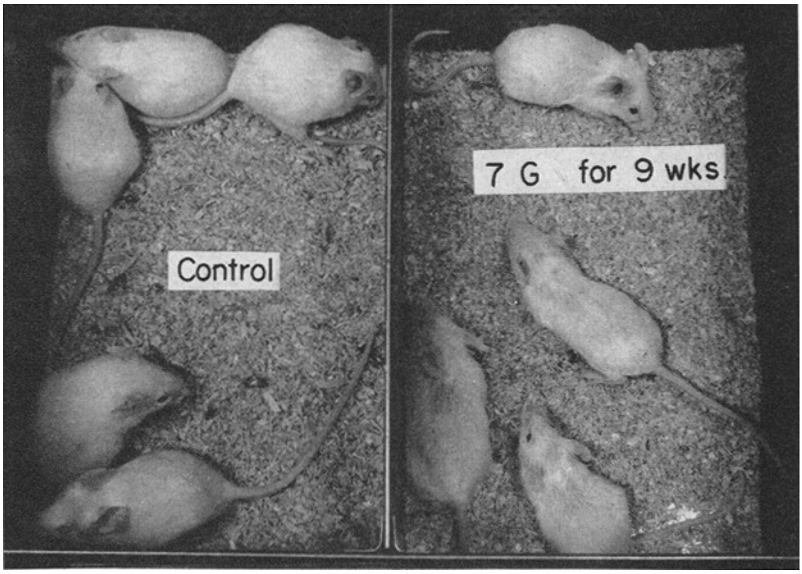

Fig. 3. These mice were five weeks of age when the experiment began. Some of the experimental mice shown here lived in the centrifuge for as long as one year. A few controls lived for slightly more than two years.

MATERIALS AND METHODS

Increased gravity was simulated by continual centrifugation in a centrifuge described elsewhere.[11] Approximately two or three male mice were housed with a comparable number of female mice in a 5 by 6-inch compartment of a cage 4 inches in height. Identical compartments were employed with both centrifuged and control cages. Mice were removed from the centrifuge for approximately ten minutes each day so that food or sawdust could be replaced and observations be performed.

Every effort was made to insure as normal an existence as possible for the mice. Bottoms of the cages were covered with ½ inch of sawdust. The mice, unrestrained except by their own artificially increased weight, were permitted to assume the same orientation to the new field as they would normally assume to the Earth's gravity. Food consisted of Purina "Laboratory

Two experiments were performed. For the results reported here, mice which had attained the age of five weeks were placed in fields of 2, 3, 5, 6, 7, 8, and 10 G throughout the remainder of their life. Other mice were also placed in 2 G when they had attained the ages of three and seven weeks. The centrifuge was operated at 105 r.p.m.

The second experiment constituted a repeat of the first with a rotational speed of 90 r.p.m. Five-week-old mice were centrifuged at 1.5, 2, 3, 5, 6 and 7 G. Other mice were placed into 2 G when three, four, and seven weeks of age. Experiments are underway with other combinations of field and age; however, such results, as well as those for female mice, are not complete at the present time.

RESULTS AND DISCUSSION

At moderate fields of acceleration, the average mouse can live almost as long as the control animal. Some mice at 7 G lived for as long as a year. At 2 G, some lived as long as two years. One mouse (female), for which data are not reported in this paper, was still alive after 30 months at 1.5 G. As would be expected, there is an obvious trend toward shorter survival time as the centrifugal intensity increases (Fig. 1).

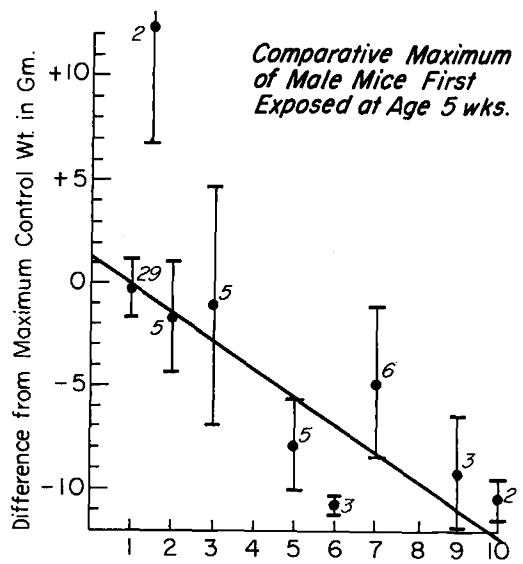

Fig. 4. Differences were measured from the control curve in Figure 5. Errors and number of mice for each point are indicated.

No detailed pathological study was pursued of the expired animals. Smith's group[9] reported that with chickens ". . . no consistent syndrome could be established for acceleration death." A cursory examination of the mice a few days before death seemed to indicate that the mice had reached "old age" at a chronologically younger age.

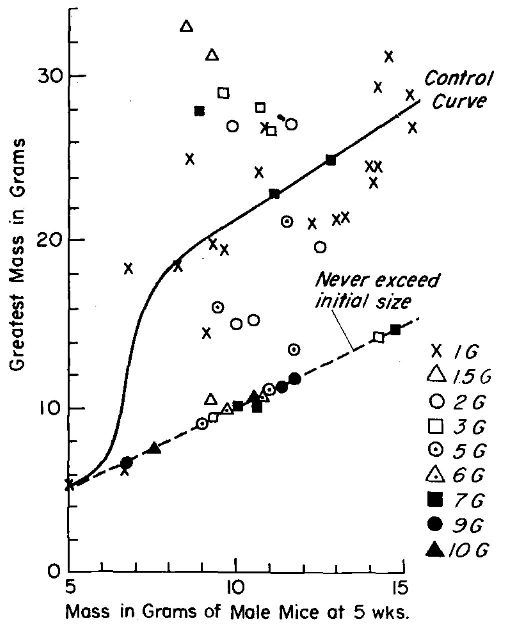

Fig. 5. Influence of size upon ability to tolerate high gravity. If an animal did not regain all of its lost body mass, initial size was assumed to be to maximal. Experimental points are for individual mice. X's indicate moving averages for control mice. The control curve was fitted by sight. Measuring differences from this curve yield values as shown in Figure 4 which are less scattered than would have been the case if all values were compared with a single control average.

Another factor which influences survival time is the period for which a mouse has been permitted to develop in the Earth's normal gravity before being placed in high gravity. Individuals which were older when placed in the centrifuge survived longer than was the case for younger animals (Fig. 2). This is contrary to what simple physical considerations and results with fly larvae would predict.[13] Also, the results for centrifuged, suckling mice are contradictory to the results reported here for weaned mice. We have observed that baby mice will grow even while their mothers, who are nursing them, are losing weight as a result of the high-G stress.

The results with older mice, however, are compatible with those of Campbell et al.[3] They observed that older rats could survive starvation for a longer period of time than do the younger ones. Centrifuged mice consume less than the normal quantity of food.[15] Actually, it is misleading to refer to the effect of initial age. Whether or not the age effect is actually due to size, maturity, adjustment to solid food, period of adjustment to one-G stress, or some other factor, has not been resolved.

In general, mice at high gravity do not attain so great a size (Figs. 2, 3 and 4). This is to be expected and consistent with other findings.[6,8,9,12] However, a moderate increase in field can sometimes cause a faster growth with fly larvae[13] and with wheat coleoptiles.[4] Perhaps the same thing has stimulated the two mice at 1.5 G to greater growth. More data are necessary for definite evidence that slight increases in gravity can stimulate the over-all growth of the mouse. However, we do know that under certain conditions the added weight load does enhance bone growth.[14]

On the basis of these results, one might predict that certain forms of life might survive for extended periods in the gravity of another planet. Perhaps it would be more appropriate to say that gravity influences the "life-span" rather than the "survival time" of an individual. The duration of the life-span is influenced by the gravitational intensity and therefore the level of the effect. Although most cells would require tens of thousands of G before they are directly affected, such fields would crush most multicellular forms in less than a second. Fields intense enough to impair circulation normally cause death within a period of seconds. With fields intense enough to impair respiratory movements, but not circulation, a period of minutes is involved. For less intense fields, an altered life-span undoubtedly results from the effects upon feeding movements,[15] digestion and metabolism in general.

SUMMARY

Developing, male, white mice were continually exposed to centrifugal fields which were known to be intense enough to hamper growth, but not cause immediate death.

Mice which had attained the age of five weeks before exposure displayed a faster mortality as field increased. Although the average mouse at 7 G lived to the age of six months, those at higher fields perished after a period of a few days.

Of mice which first attained an age of three, four, five or seven weeks before exposure, older ones displayed the best survival.

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