The study of aging in man is difficult because of the lack of genetic and environmental control. In the rat these factors can be controlled and the findings in animals can supply information and valuable clues to human gerontology. On this premise our rat colony was established more than 10 years ago and over 50 generations of inbred animals have been raised under conditions particularly favorable for long survival.

In connection with our studies on aging it seemed advisable to extend our observations so as to include studies on growth in relation to longevity and senescence. Accordingly data on body weight, body length, and tibia length were obtained under carefully controlled conditions in rats from weaning through maturity and old age. Parallel studies on the histology of the proximal tibial epiphysis were also made to determine whether epiphyseal non-union, a phenomenon peculiar to the rat, was significant with respect to potential growth in old animals. These findings together with observations on the influence of chronic disease on growth are described in the present paper.

Lung infection is endemic and is the major cause of death in most rat colonies. The disease seriously affects the validity of experimental results and especially hampers studies on growth and longevity. By a process of elimination and selection described later, pulmonary infection has been largely excluded from our colony so that data have been derived wholly from animals with uninfected lungs. Mortality has been due entirely to other diseases.

Rat Selection.—Data were obtained from a total of 858 Sprague-Dawley rats, 543 males and 315 females. Details as to numbers at different age levels and as to health status are given in tables 1-4. The animals fell into 2 categories. The first consisted of 133 males and 138 females which continued to gain weight until they were sacrificed (tables 1 and 2). These rats showed at autopsy no discernible gross or microscopic lesions. Up to about 170 days, random choice was employed since the incidence of lesions until this age was negligible. Later, when pathologic changes began to appear, selection became necessary and with advancing age it was used to a greater and greater extent as the frequency of lesions increased. After 681 days in males and after 801 days in females, the number of disease-free animals was too small to obtain significant mean values for further growth studies. Since tissue sampling was restricted to relatively small sections of organs, it is possible that some rats classified as free from lesions actually had undetected lesions. In such cases, however, the changes were not far enough advanced to interfere with steady weight gain.

The second category comprised 410 males and 177 females; all had lesions of moderate or marked severity and had lost weight. Most of these animals were over 700 days old but some, especially males, were younger (tables 3 and 4). No selection was made in this category, excepting a small number of animals with pulmonary infection which were excluded.

In the study by Simms and Berg (4) on the accumulation and progression of lesions in relation to age, all of the data were based on findings in rats chosen at random. The present investigation differed in that after the onset of disease, selection was employed in order to obtain data on animals free from lesions. In both studies classification of lesions according to type and severity was the same.
quiet atmosphere, gentle handling, and avoidance of stresses were emphasized in the management of the colony.

Control of Pulmonary Infection.—Floors, walls, and attendants' uniforms and gloves were kept clean; dust from pellets was sifted and discarded. Cages and water bottles were sterilized at regular intervals. Visitors were kept at a minimum and were required to wear gowns.

The program to control lung infection was started 6 years ago and initially all rats with the following signs or symptoms were removed from the colony: 1) bloody nasal discharge; 2) rapid, labored respiration; 3) rhonchi due to sticky tracheobronchial secretions; 4) male rats up to 550 days old and females up to 750 days old having no obvious respiratory symptoms but either failing to gain weight or losing weight. The symptom-free survivors were watched closely for evidence of early involvement and such rats were promptly eliminated. Particularly important was the removal of infected breeders. Rigorous adherence to this regime reduced the incidence of chronic and acute lung infection from an over-all figure of more than 50 per cent to 2 per cent for the chronic type and to 4 per cent for the acute type. This low incidence has been maintained for the past 5 years. The chronic form is characterized by suppurative bronchiectases, bronchiectatic abscesses, and pneumonitis. The acute type is a fulminating, rapidly fatal, hemorrhagic, necrotizing pneumonitis, often with abscess formation. None of the animals included in this study had infected lungs.

Histologic Examinations.—Apparently healthy animals used to establish growth curve; in the absence of lesions were killed at different ages by intraperitoneal injection of nembutal. The balance of the rats either completed their life span or were killed by nembutal. The program to control lung infection was started 6 years ago and initially all rats with the following signs or symptoms were removed from the colony: 1) bloody nasal discharge; 2) rapid, labored respiration; 3) rhonchi due to sticky tracheobronchial secretions; 4) male rats up to 550 days old and females up to 750 days old having no obvious respiratory symptoms but either failing to gain weight or losing weight. The symptom-free survivors were watched closely for evidence of early involvement and such rats were promptly eliminated. Particularly important was the removal of infected breeders. Rigorous adherence to this regime reduced the incidence of chronic and acute lung infection from an over-all figure of more than 50 per cent to 2 per cent for the chronic type and to 4 per cent for the acute type. This low incidence has been maintained for the past 5 years. The chronic form is characterized by suppurative bronchiectases, bronchiectatic abscesses, and pneumonitis. The acute type is a fulminating, rapidly fatal, hemorrhagic, necrotizing pneumonitis, often with abscess formation. None of the animals included in this study had infected lungs.

Histologic Examinations.—Apparently healthy animals used to establish growth curve; in the absence of lesions were killed at different ages by intraperitoneal injection of nembutal. The balance of the rats either completed their life span or were killed by nembutal when they were moribund or had hind leg paralysis (2). Complete autopsies were performed immediately after death; animals dying sponta-
males and decreased progressively with advancing age in the female. Males reached their maximum weights, increments at corresponding ages were greater in males free from lesions than in females and decreased progressively with advancing age (tables 1, 2 and figs. 1, 2). Weight increases continued to a later age in the female than in the male because disease set in later in the female. Males reached their maximum weights (mean of 486 Gm.) at 522 days and females attained their highest weights (mean of 279 Gm.) at 801 days. The subsequent course of the weight curves followed a plateau and then a decline, a manifestation of advancing disease process. In the younger animals in which lesions had developed sooner there was a corresponding earlier break in the weight curves. Relatively high weights were found in rats with tumors or fluid accumulations but the net values after removal of the tumor or fluid were due partly to deposition of fat in the subcutaneous tissue. 

Measurements.—Body weights were terminal values. Tibia length was determined by precision calipers and was the distance between the spines of the tibial head and the medial malleolus. Body length was the calipered measurement from the tip of the nose to the anus.

RESULTS

Body Weight.—Starting at equal weaning weights, increments at corresponding ages were greater in males than in females and decreased progressively with advancing age (tables 1, 2 and figs. 1, 2). Weight increases continued to a later age in the female than in the male because disease set in later in the female. Males reached their maximum

### Table 3. Measurements of Male Rats Having Lesions.

<table>
<thead>
<tr>
<th>Age (Days)</th>
<th>Tibia Length (cm.)</th>
<th>Body Length (cm.)</th>
<th>Body Weight (Gm.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Range</td>
<td>No.</td>
</tr>
<tr>
<td>469</td>
<td>417-496</td>
<td>12</td>
<td>4.55</td>
</tr>
<tr>
<td>570</td>
<td>500-507</td>
<td>34</td>
<td>4.56</td>
</tr>
<tr>
<td>655</td>
<td>602-699</td>
<td>76</td>
<td>4.57</td>
</tr>
<tr>
<td>747</td>
<td>700-798</td>
<td>84</td>
<td>4.57</td>
</tr>
<tr>
<td>854</td>
<td>802-898</td>
<td>86</td>
<td>4.56</td>
</tr>
<tr>
<td>940</td>
<td>900-998</td>
<td>76</td>
<td>4.55</td>
</tr>
<tr>
<td>1038</td>
<td>1002-1090</td>
<td>38</td>
<td>4.51</td>
</tr>
<tr>
<td>1124</td>
<td>1110-1142</td>
<td>4</td>
<td>4.52</td>
</tr>
</tbody>
</table>

Totals . . . . . | 410 | 365 |

### Table 4. Measurements of Female Rats Having Lesions.

<table>
<thead>
<tr>
<th>Age (Days)</th>
<th>Tibia Length (cm.)</th>
<th>Body Length (cm.)</th>
<th>Body Weight (Gm.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Range</td>
<td>No.</td>
</tr>
<tr>
<td>544</td>
<td>501-599</td>
<td>12</td>
<td>3.98</td>
</tr>
<tr>
<td>643</td>
<td>601-692</td>
<td>15</td>
<td>3.97</td>
</tr>
<tr>
<td>744</td>
<td>709-796</td>
<td>20</td>
<td>3.98</td>
</tr>
<tr>
<td>860</td>
<td>805-899</td>
<td>38</td>
<td>3.98</td>
</tr>
<tr>
<td>953</td>
<td>902-999</td>
<td>38</td>
<td>4.01</td>
</tr>
<tr>
<td>1061</td>
<td>1003-1098</td>
<td>39</td>
<td>3.97</td>
</tr>
<tr>
<td>1143</td>
<td>1104-1190</td>
<td>12</td>
<td>3.90</td>
</tr>
<tr>
<td>1200</td>
<td>1210-1246</td>
<td>3</td>
<td>4.04</td>
</tr>
</tbody>
</table>

Totals . . . . . | 177 | 150 |

Mean values after removal of the tumor or fluid were due partly to deposition of fat in the subcutaneous tissue. 

Specimens were taken from all tissues and after fixation in Zenker's fluid were imbedded in paraffin. Sections were stained with hematoxylin and eosin for histologic study. The proximal epiphysis of the tibia was prepared for examination by preliminary decalcification in 5 per cent nitric acid solution.

Specimens were taken from all tissues and after fixation in Zenker's fluid were imbedded in paraffin. Sections were stained with hematoxylin and eosin for histologic study. The proximal epiphysis of the tibia was prepared for examination by preliminary decalcification in 5 per cent nitric acid solution. 

RESULTS

Body Weight.—Starting at equal weaning weights, increments at corresponding ages were greater in males than in females and decreased progressively with advancing age (tables 1, 2 and figs. 1, 2). Weight increases continued to a later age in the female than in the male because disease set in later in the female. Males reached their maximum weights (mean of 486 Gm.) at 522 days and females attained their highest weights (mean of 279 Gm.) at 801 days. The subsequent course of the weight curves followed a plateau and then a decline, a manifestation of advancing disease process. In the younger animals in which lesions had developed sooner there was a corresponding earlier break in the weight curves. Relatively high weights were found in rats with tumors or fluid accumulations but the net values after removal of the tumor or fluid was low. The commonest pathologic conditions occurring in our rat strain were chronic nephrosis and glomerulonephritis, periarthritis, myocardial degeneration and fibrosis, skeletal muscle dystrophy, and certain tumors, viz., adenoma of the anterior lobe of the pituitary, phaeochromocytoma of the adrenal medulla, adenoma of the thyroid, and fibroadenoma of the breast in females.

Body weight increased rapidly up to about 170 days after which the rate of increase diminished sharply. Increments with advancing age are due partly to deposition of fat in the sub-
cutaneous tissue, in the retroperitoneal and perirenal spaces, in the mesenteries of the spermatic cord, of the ovary, of the uterus, and of the intestine and in the interscapular area. Whether the accumulation of fat in the mature rat can be termed normal is questionable. Zucker and associates (6) have an affirmative opinion based on the finding that rate of fat deposition does not disturb the straight line relation between log body weight plotted against reciprocal of age. Nevertheless the fact remains that fat accumulation is caused by an imbalance between energy requirement and food intake and to this extent is abnormal. Certainly cage confinement restricts spontaneous activity more and more as an animal grows and thus contributes to fat deposition; unlimited food intake furthers it. It is plausible that regulation of food intake according to energy needs will reduce fat formation, will diminish rate and extent of body weight increments, and body weight curves will be nearly flat after 170 days.

**Tibia Length**—At weaning, although body weights were equal, the tibia was longer in males than in females and continued to grow at a faster rate in males. Between 100 and 681 days in males there was a 12 per cent increment in tibia length while in females the increase from 108 to 801 days was only 7 per cent. Until about 170 days in both sexes rate of growth of the tibia was rapid and then declined abruptly. Later, increments became extremely small but were found to be statistically significant when a sufficient interval elapsed between measurements. For example, in males the P value was 0.001 for the difference between the maximum mean length of 4.61 cm. at 681 days and the mean length of 4.50 cm. at 328 days; for females, the P value was 0.001 in comparing the maximum mean length of 3.99 cm. at 801 days with the mean length of 3.93 cm. at 357 days. Evans and coworkers (1, 5) found no appreciable change in tibia length in female rats after approximately 120 days. After peak tibia length was reached, time intervals of later measurements were too short to ascertain if further statistically significant growth occurred. The lower values for tibia length in male rats over 1,000 days old could be explained on the basis of shrinkage of bone matrix, atrophy of articular cartilage, or osteoporosis. Another explanation could be unintentional selective breeding giving larger animals among the younger age groups.

Of particular interest was the finding that mean tibia lengths were nearly the same in rats.
Fig. 3. 70-day-old male. Osteogenesis is active. Numerous primary and secondary bony trabeculae extend from diaphyseal side of cartilage plate which is wide and even.

Fig. 4. 180-day-old female. Osteogenesis is less active. Fewer trabeculae are present and continuity between primary and secondary ones is broken. Cartilage plate is narrower and uneven.

Fig. 5. 316-day-old female. Osteogenetic activity is reduced further. Number of primary trabeculae is greatly diminished and only scattered remnants of secondary trabeculae remain. Irregularity and shrinkage of cartilage plate is more pronounced.

Fig. 6. 790-day-old male. Osteogenesis is minimal. Several long, thin trabeculae extend from cartilage plate, which is very irregular.

Fig. 7. 1071-day-old female. No evidence of osteogenesis is present. Disc is reduced to a narrow, irregular band of cartilage enclosed in bony plates to which a few, thick primary trabeculae are attached.

Fig. 8. 1054-day-old male. Disc is relatively wide but uneven. On diaphyseal side of cartilage are blunt ends of trabeculae which have largely disappeared.

Fig. 9. 1133-day-old female. Marked disarrangement and degeneration of cartilage cells. Disc persists but incased in bony plates. A few remnants of trabeculae remain.

Figures 3 through 9 show typical changes, at different ages, in the proximal tibial epiphysis of the rat. All 10X. Hematoxylin eosin.
with advanced lesions and with weight loss as in those free from lesions. For instance, the mean tibia length for males free from lesions was 4.58 cm. at 522 days while in their counterparts with well developed lesions and weight loss, it was 4.56 cm. at 579 days. Similarly in females the tibias of animals free from lesions averaged 3.96 cm. at 585 days as compared to 3.98 cm. at 544 days for those with lesions. In fact, the maximum female tibia measurement of 4.01 cm. was found at 952 days in rats with disease and considerable weight loss.

**Body Length.** — Rate of increase of body length closely paralleled rate of tibia growth. For males with no detectable lesions a maximum mean body length of 25.0 cm. was found at 622 days; the corresponding value for females was 21.2 cm. at 801 days. In females measurements were about the same in animals with or without lesions but in males body lengths were shorter in those with lesions. This difference was probably due to the greater loss of subcutaneous perineal fat in the male.

*The Proximal Tibial Epiphysis.* — In general the order of closure of the skeletal epiphyses in the rat is the same as in other mammals (5). Most of the epiphyses in the rat have united by 120 days. However, certain epiphyses fail to close even in old age. At least one epiphysis in each of the major long bones and the epiphyses of the caudal vertebrae are patent into senescence.

In figures 3 to 9 are shown the changes that are found in the proximal tibial epiphysis of rats from 70 days to over 1100 days old. These are presented as typical alterations with advancing age. Some variation in the appearance of the epiphysis is found in old animals, but there is no evidence of osteogenetic activity.

During early life when growth is rapid the cartilage plate of the proximal tibial epiphysis is wide, and numerous long, thin, interlacing, newly formed bony trabeculae are found on the diaphyseal side of the disc. By about 170 days when increments in tibia length diminish markedly there is corresponding histologic evidence of decreased osteogenesis at the epiphysis. The width of the cartilage is decreased, the margins are somewhat irregular, and the number of new trabeculae is reduced. At about 300 days there are further signs of diminishing osteogenesis. Narrowing and irregularity of the cartilage are greater and trabeculae are fewer. With advancing age regressive changes in the epiphysis become more pronounced. The cartilage becomes increasingly narrow and irregular and new bone formation is minimal. In very old rats over 1,000 days of age a zone of cartilage persists but it is incased in bony plates. A few stubby trabeculae remain attached to the disc but there is no evidence of active osteogenesis. In animals over 1100 days old the epiphysis is still open.

**Comparative Growth Rates and the Principle of Heterogony**

Huxley (3) demonstrated that the ratio of relative growth rates between any 2 parts of the body, or a part of the body and the whole, remains constant. He also showed that the change in the log of organ weight or organ dimensions at a given time is proportional to the change in the log of body weight or body dimensions during the same time. To this relation Huxley applied the term heterogony.
Applying Huxley's principle to our own material but using linear instead of logarithmic scales, the measurements of male and female rats free from lesions were plotted to determine the relation of tibia length and body length growth rates. The points fall on a relatively straight line common to both sexes, except for a slight downward concavity for higher values in females (fig. 10). When the linear plot is transferred to a logarithmic scale the resultant straight line has the slope of unity and conforms with Huxley's principle of heterogony.

A similar plot of tibia length and cube root of body weight again shows a linear relation with points for both sexes falling on a single line with a slight downward concavity at higher values for females (fig. 11). These data are also consistent with Huxley's principle, and with Zucker and Zucker's finding of a constant relation between femur bone ash weight and body weight (6).

It is of interest that even though males and females have quite different growth rates, the data of both sexes in the 2 graphs fall on one line, demonstrating that both sexes have the same proportionality constant when 2 body dimensions are compared.

DISCUSSION

As a result of controlling lung infection, the average longevity of rats in our colony has been increased considerably, in males to about 750 days, and in females to about 900 days. However, the maximum life span has not been lengthened beyond 1200 days in the male and 1300 days in the female, because the diseases which develop with advancing age, and are the chief causes of death, continue to operate as limiting factors. A similar situation exists in man. With the introduction of new therapeutic agents such as antibiotics and insulin, life expectancy has been increased but the maximum life span has not changed due to the fact that the toll from the so-called degenerative diseases of old age has not been altered.

In the rat, chronic pathologic conditions appear while skeletal growth is going on, but in man chronic disease develops largely after skeletal growth has stopped (the last epiphysis closes at about 20 years of age). Increments in skeletal size continue for about 60 per cent of the rat's life span while in man the period of skeletal growth occupies less than 30 per cent of the life span. The principal lesions found in senescent rats are in the categories of cardiovascular-renal lesions and neoplasms, and they involve the same tissues that are affected in aging man.

On a histologic basis, persistence of certain epiphyseal cartilages in very old rats seems to indicate that skeletal growth has not yet ceased. Our findings support the generally accepted idea that the epiphyseal disc does not unite even in the senescent animal. But the histologic appearance in very old rats is such as to indicate that further measurable growth is very unlikely.

It is a moot question whether or not the pathologic changes occurring in later life are degenerative in the sense that they represent a breakdown of intrinsic biologic systems within tissue cells. Whether or not these changes are inevitable with time is another debatable question. An increased susceptibility to disease has been found with advancing age (4). For a better understanding of the aging process, it would be helpful if we could extend the life span of our experimental animals by preventing or eradicating the diseases that occur in the older animals. An effort in this direction is our present study on muscular dystrophy, the most frequent terminal cause of death of old rats in our colony. The skeletal muscle lesions resemble the changes resulting from experimental vitamin E deficiency (2) and on this basis large doses of alpha tocopherol have been given to a group of rats to determine if the spontaneous muscle disease can be prevented or cured.

The female rat is superior to the male with respect to vigor, to longevity, to resistance to disease, and to length of period of continued growth. Experiments in progress are designed to determine whether these differences are related to the smaller size of the female or to other factors, such as hormones. These studies should also help to elucidate whether maximum growth is optimum growth as considered in relation to susceptibility to disease and to longevity.

SUMMARY

Procedures that have been successful in controlling lung infection in our rat colony have been described. Rates of growth for both sexes, as determined by body weight, body
length, and tibia length have been established under standardized conditions from weaning through maturity and old age. Data are given for animals free from lesions and for animals with chronic disease. Linear plots of tibia length against body length and of tibia length against the cube root of body weight give points for both sexes falling on a straight line. These data are in accord with Huxley's principle of heterogony. Histologic studies of the proximal tibial epiphysis show non-union even in senescence but there is no evidence of continued osteogenesis into old age. These standardized patterns of growth can be used as the basis for comparative studies on the effects of growth retardation in relation to prolongation of the life span.

REFERENCES