

5. The data indicate that ferritin is involved in the phenomenon of iron absorption through the intestine of the horse, and that the lymphatic system is concerned with iron absorption by some process in which ferritin also plays a rôle.

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## Collagen Content of Guinea Pig Tissues. (18122)

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A chemical method has been described by which collagen, the major fibrous component of connective tissue, can be measured quantitatively in tissues(1). This technic has been used to study the effects of physiologic and pathologic processes on the formation, distribution and destruction of collagen(1-13). During the course of an investigation of the effect of ascorbic acid deficiency on the collagen content of guinea pig tissues(6), the necessity for a more detailed study of the

normal pattern of connective tissue distribution in the body became more evident. Therefore, this study was undertaken.

**Experimental.** One hundred and eight male guinea pigs weighing between 60 and 1000 g were selected from an inbred AMC strain at this laboratory. The animals were maintained on a diet of Derwood Rabbit Chow pellets and fresh greens daily *ad libitum*. They were weaned at approximately 200 g body weight. The guinea pigs were sacrificed with a blow to the back of the skull, which occasioned a variable amount of bleeding

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RELATION OF ORGAN WEIGHT TO MEAN BODY WEIGHT OF MALE GUINEA PIGS

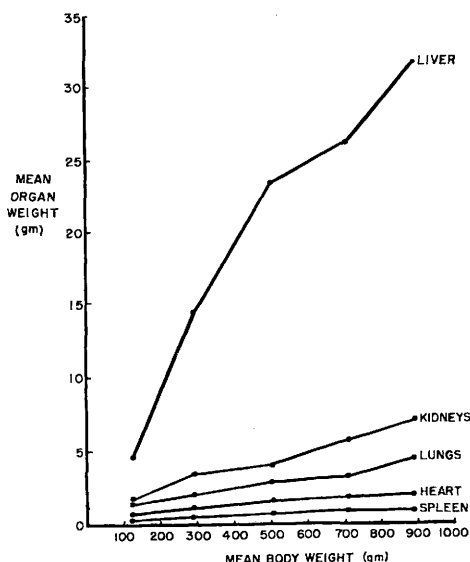


FIG. 1.

TABLE I. Collagen Content of Wet Tissues.

| Group (g) | No. of animals | Mean wt (g)* | Lungs (%)* | Liver (%)* | Kidneys (%)* | Spleen (%)* | Heart (%)* | Skeletal muscle (%)* |
|-----------|----------------|--------------|------------|------------|--------------|-------------|------------|----------------------|
| 60-199    | 11             | 121 ± 14     | 1.01 ± .10 | .36 ± .06  | .47 ± .06    | 1.21 ± .32  | .80 ± .13  | .96 ± .12            |
| 200-399   | 17             | 296 ± 13     | 1.45 ± .08 | .63 ± .08  | .62 ± .05    | .74 ± .08   | 1.06 ± .06 | .89 ± .04            |
| 400-599   | 10             | 501 ± 19     | 1.47 ± .09 | .47 ± .07  | .60 ± .05    | .54 ± .07   | 1.18 ± .05 | 1.00 ± .08           |
| 600-799   | 10             | 706 ± 13     | 1.79 ± .12 | .39 ± .03  | .56 ± .03    | .55 ± .08   | 1.04 ± .05 | .98 ± .06            |
| 800-999   | 10             | 888 ± 13     | 1.67 ± .12 | .37 ± .04  | .61 ± .04    | .50 ± .05   | 1.06 ± .06 | 1.25 ± .09           |

\* Includes stand. error of the mean.

from the cranial orifices. They were weighed and dissected immediately. Samples of skeletal muscle, heart, lungs, liver, kidneys and spleen were removed in a manner described previously(6). In 58 animals, quantitative chemical measurements of the collagen content of the various tissues were made according to the method of Lowry *et al.*(1). If the weight of the individual organ was 4 g or less, the entire organ was used for the chemical analysis. In all instances the heart and spleen were treated in this manner. If the organ was larger than 4 g, it was weighed to the nearest 0.1 g and a sample taken for the chemical procedure. Liver and skeletal muscle fell into this category. Depending on the weights of the lungs and kidneys, analyses were conducted either on the single organ or on the pair of organs. The proportion of water in the tissues of the remaining guinea pigs was measured. Approximately 1 g of tissue was minced, weighed and desiccated to constant weight.

The relation of mean organ weight to mean body weight is illustrated in Fig. 1. The 58 animals were divided by weight into 5 groups. The rate of growth was characteristic for each organ, the liver growing most rapidly and the spleen least rapidly.

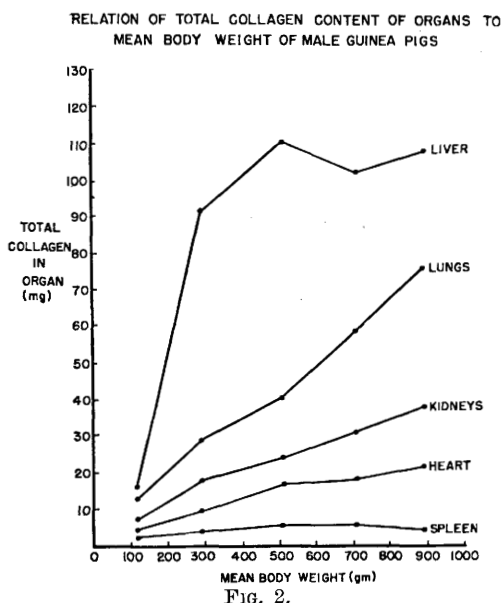
The collagen content of the various tissues on a wet weight basis is listed in Table I. The amount of tissue collagen varied with the type and size of the organ. The lungs contained the largest, and the liver the smallest concentration of collagen. The changes of collagen content with age were not uniform in all the tissues. One pattern was exemplified by the lung, in which a progressive increase occurred with increase of body weight. The proportion of connective tissue

in skeletal muscle remained approximately constant except for a significant increase in the oldest group. On the other hand, there was an initial rise of the concentration of collagen in the kidneys and heart, after which the collagen level remained unaltered. In the liver the proportion of collagen increased at first, attained a maximum and then decreased. The spleen exhibited the most unusual change with age; the concentration of collagen decreased progressively as the animal increased in size. When values for collagen content of wet tissues were corrected for water content, as obtained from 50 guinea pigs of comparable weight, the derived data of Table II were calculated. Expression of collagen on a dry weight basis did not alter significantly the findings described above. The absolute amounts of collagen in the tissues are diagrammed in Fig. 2. The values are the product of the organ weight and the collagen concentration. There was an increase of the amount of collagen in each organ with somatic growth, despite the fluctuations of the proportions of collagen in the tissue.

**Discussion.** Several factors may be considered in evaluating these results. For a population of healthy guinea pigs, body

TABLE II. Collagen Content of Dry Tissues.

| Group (g) | Lungs (%) | Liver (%) | Kidneys (%) | Spleen (%) | Heart (%) | Skeletal muscle (%) |
|-----------|-----------|-----------|-------------|------------|-----------|---------------------|
| 60-199    | 5.05      | 1.21      | 2.41        | 5.53       | 3.94      | 4.44                |
| 200-399   | 7.59      | 2.23      | 2.97        | 3.46       | 5.58      | 4.16                |
| 400-599   | 7.42      | 1.64      | 2.83        | 2.51       | 6.21      | 4.57                |
| 600-799   | 8.91      | 1.32      | 2.55        | 2.49       | 5.42      | 4.28                |
| 800-999   | 8.15      | 1.28      | 2.81        | 2.25       | 5.44      | 5.46                |



weight has been satisfactorily used as the criterion of age(14). In this investigation the effect of sex was not explored; only male animals were studied. It was not possible to determine simultaneously both collagen and water content of the tissues in the same animals. Therefore, 2 sets of data were obtained and pooled in order to calculate the collagen concentrations on a dry weight basis. Although it would have been more desirable to have the data from the same animals, these results are nevertheless valid. The other constituents of the tissues were not measured chemically. Relative changes in the proportions of connective tissue may be occasioned by variations of the other constituents. As growth proceeds, there is increased accumulation of connective tissue in the different organs.

The amount of collagen in guinea pig liver, kidney, skeletal muscle and spleen does not

differ greatly from the corresponding organs of the human and rat(1,3-5,7,9). The values for the guinea pig heart could not be compared with reported values for rat(9) or human(12). In our study, the entire organ, including valves, chordae tendineae and auricles were examined *in toto*; in the other studies, only the cardiac musculature was examined. No previously published values for the normal collagen content of lung have been found.

The general statement that progressive fibrosis of tissues occurs with aging should be qualified. In these experiments, older tissues contained more collagen than younger ones, with the exception of the spleen. Lowry *et al.*(7) failed to find a correlation between the age and the connective tissue content of rat livers. In contrast, there was a definite alteration of the liver collagen content with age in the guinea pig. Increased fibrosis of rat skeletal muscle(9) and renal cortex(7) in older animals has been reported. Our findings confirmed the former but failed to substantiate the latter in the guinea pig whole kidney.

**Summary.** 1. Chemical measurements were made of the collagen content of the lungs, liver, kidneys, spleen, heart and skeletal muscle of 58 guinea pigs weighing between 60 and 1000 g. 2. The concentration of connective tissue components varied with different tissues; the lungs contained the greatest proportion, and the liver the least proportion, of collagen. 3. Tissues from older animals usually contained a higher per cent collagen than comparable younger tissues, except for the spleen where the converse was true. 4. The values obtained for guinea pig tissues were in essential agreement with previously reported figures for other species.

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