14698

Distribution of Bone Marrow, Bone, and Bone-Ash in Rabbits.

ALBERT A. DIETZ. (Introduced by Bernhard Steinberg.)

From the Toledo Hospital Institute of Medical Research, Toledo, O.

Although bone marrow is an important organ, few quantitative studies of it have been made. In the investigations being conducted in this laboratory, a knowledge of distribution of bone marrow in rabbits became desirable. Nye, using a modification of the method of Wetzel,² found the total marrow in 2 rabbits to be equal to 2.00 and 2.59% of the body weight. Similar results have been obtained for dogs³ and newborn infants.⁴ In the latter case, one-third of the marrow was found in the skull. The above workers removed the soft tissues by maceration, extracted the fat of the whole bone with ether, and filled bone marrow cavities with agar, gelatin, or Woods metal. The volume of the whole bone was then measured by displacement of water and the volume of the bony structure calculated from the weight and density of bone. bone marrow volume constituted the difference between these two values. This method represented the bony structure to contain marrow since maceration and extraction leave it porous. It also assumed that the medullary cavities were filled to the same extent that they previously contained marrow. errors are probably compensated when the marrow of the whole animal is calculated but for individual bones the results are erroneous.

Mechanik⁵ determined the bone marrow content of individual bones of 13 cadavers. He considered the bone marrow to be the difference in weight of fresh and macerated bone, after correcting for the loss of bone tissue. He found the total marrow to equal

3.4-6.5 (average 4.6) % of the body weight, and recorded the bone marrow content of individual bones in numerous tables.

Neither of the above methods can be applied readily to the determination of the marrow content of individual bones of rabbits. Advantage was taken of the large difference in the ash content of bone and bone marrow. From the ash analyses of the whole bone, marrow-free bone and bone-free marrow, the quantity of bone and bone marrow present can be calculated. Since the weight of the whole bone, a, equals the sum of the weights of the marrow, x, and the marrow-free bone, y, and the weight of the ash of the whole bone, b, is equal to the weight of the ash of these components, 2 equations in 2 unknowns are obtained: x + y = a, and cx + dy = b. In the latter c and d are the grams of ash per gram of marrow and marrow-free bone, respectively. Solving these simultaneous equations for x, one obtains:

$$x$$
 bone marrow $=\frac{ad-b}{d-c}$ (I)

The bone tissue then is the difference between the weights of the whole bone and the bone marrow, or its weight can be obtained directly by solving the equations for y. The ash of the marrow-free bone was determined for as many bones as lent themselves to the method. Because of the low ash content of the bone marrow, representative analyses were used in the calculations. Its ash content can be neglected without introducing a significant error, i.e., c = 0. Theoretically, one can use constituents other than ash for the determination of the marrow content of individual bones, but the differences in the analyses of bone and marrow are not so marked nor so uniform.6 Since the bone and bone-ash are determined simultaneously with the bone marrow, their distribution is included in the tables.

¹ Nye, R. N., Proc. Soc. Exp. Biol. and Med., 1931, 29, 34.

² Wetzel, G., Arch. f. Entwcklngsmechn. d. Organ., 1910, **30**, 507.

³ Fairman, E., and Whipple, G. H., Am. J. Physiol., 1933, **104**, 352.

⁴ Töppich, G., Arch. Anat., 1914, 9.

⁵ Mechanik, N., Z. f. Anat. u. Entwcklngsgesch., 1926, **79**, 58.

⁶ Krause, R. F., J. Biol. Chem., 1943, 149, 395.

TABLE I.
Distribution of Bone Marrow, Bone, and Bone-ash in 5 New Zealand Rabbits.

	Marrow*		Bone	Bone*		Whole bone*		Bone-ash*	
	Range, %	Avg, %	Range, %	Avg, %	Range, %	Avg, %	Range, %	Avg, %	
Ossa Coxæ	8.32-10.04	9.19	5.57- 7.12	6.61	7.44- 7.89	7.61	6.32- 6.90	6.65	
Femora	14.08-19.87	17.11	9.39 - 10.43	9.95	11.34-13.86	12.57	10.51-11.53	11.06	
Tibiæ	$9.73 \cdot 12.61$	11.54	8.86- 9.71	9.18	9.96 - 10.42	10.04	9.61 - 10.36	9.94	
Fibulæ	0.26 - 0.41	0.34	0.30 - 0.42	0.36	0.34 - 0.39	0.36	0.34- 0.42	0.39	
Tarsi and Patellæ	3.32- 4.18	3.58	3.27- 3.69	3.43	3.29- 3.68	3.48	3.16- 3.82	3.45	
Metatarsals	2.34-4.10	3.15	3.65 - 3.83	3.75	3.20- 3.93	3.53	3.47- 3.88		
Other bones of pedes	1.51- 2.11	1.74	2.14- 2.48	2.22	1.88- 2.17	2.03	2.06- 2.31	2.22	
Total hind legs	44.08-49.96	46.65	34.07-36.64	35.51	38.21-40.18	39.61	36.19-38.72	37.43	
Scapulæ	2.58- 2.94	2.76	2.68- 3.52	3.18	2,80- 3,28	3.04	2.84- 3.51	3.16	
Humeri	7.41- 9.18	8.46	4.48- 5.67	5.31	5.91- 7.07	6.47	5.36- 6.20	5.75	
Ulnæ	2.12- 2.90	2.57	2.68- 3.17	2.89	2.68- 2.88	2.77	2.81- 3.11	2.91	
Radii	1.57- 1.99		1.80- 2.06	1.95	1.75- 2.03	1.91	1.86- 2.07	1.96	
Manus	1.54- 2.56		2.30- 2.92	2.62	2.18- 2.77	2.39	2.23- 2.70		
Total fore legs	16.69-18.47	17.67	14.92-16.71	15.95	16.04-17.05	$\overline{16.59}$	15.55-17.15	$\frac{-}{16.31}$	
Ribs	4.47- 6.08	5.25	5.07- 6.21	5.82	4.82- 6.10	5.62	4.63- 5.65	5.34	
Sternum and clavicul	æ 0.72- 0.94		0.38- 0.54	0.48	0.58- 0.69	0.62	0.36- 0.50		
Total chest	5.32- 6.95	6.11	5.45- 6.75	6.31	5.40- 6.78	6.24	4.99- 6.15	5.80	
Skull	5.55- 8.27	7.14	13.98-17.35	15.44	11.41-13.24	12.37	13.00-16.03	14.75	
Mandibulæ	0.95 - 2.15	1.45	5.19 - 7.79	6.08	3.87- 5.45	4.36	5.38- 7.86	6.23	
Hyoid	0.08- 0.13	0.09	0.04- 0.06	0.05	0.05- 0.07	0.06	0.04- 0.11	0.08	
Total head	6.73- 9.92	8.68	19.49-25.20	21.57	15.33-18.76	16.79	18.70-24.00	21.06	
Vertebræ, Cervical	2.21- 3.72	2.93	3.80- 4.69	4.16	3.30- 4.15	3.72	3.52- 4.30	3.81	
'' Thoracie	5.08- 6.33		5.41- 6.00	5.61	5.29- 5.91	5.61	4.84-5.48		
'' Lumbar	8.26- 9.63		8.29- 8.96	8.72	8.28- 9.25	8.83	7.90- 8.66	8.36	
'' Sacral	2.05- 3.08		1.33- 1.77	1.52	1.59- 2.20	1.90	1.26- 1.69		
'' Caudal	0.62- 1.16		0.57 - 0.72	0.66	0.59- 0.90	0.72	0.54- 0.71		
Total vertebræ	19.27-23.28	20.89	20.26-21.41	20.66	19.89-21.59	20.78	18.87-20.00	19.40	

^{* %} of total.

Five adult New Zealand Experiments. rabbits of different stocks were used. jugular veins were severed and the animals bled to remove as much blood from the marrow as possible. The dead animals were stored in a refrigerator, and only a few bones were removed at a time for dissection. prevent loss of moisture, the bones were kept moist with saline during dissection. In cases of duplicating bones, those from the right side were used for analyses of the whole bone. Marrow-free portions of bone and bone-free marrow from the corresponding bones of the left side were analyzed separately. Where there were no corresponding bones, e.g., vertebræ, a marrow-free portion of the bone was analyzed separately. The remaining portion was also ashed, and the sum of the ash of the two parts gave the ash content of the whole bone. For a few bones, *i.e.*, manus, caudal vertebræ, sternum, the analysis of closely related marrow-free bone had to be used in the calculations. All bones, especially those of the head were carefully dissected to remove all non-marrow soft tissue.

The samples were weighed, dried at 110°C, and ashed at 700-800°C. The marrow content was calculated from equation (I). The ash of the marrow was found to be 0.4 to 0.6% and 0.5% was used for the calculations. The results of the calculations are shown in Table I.

Results. These experiments show that the bone marrow of adult rabbits constitutes about one-third of the weight of the skeleton, or about 2.2% of the total body weight. These

TABLE II.									
Amount of Bone	Marrow and	Other	Organs i	n Individual	Rabbits.				

·			Rabbit No.					
Organ		1 0	2 8	3 &	4 8	5 Q	Avg	Avg dev.
Body wt	g	1740	2050	2100	3100	3220		,
Liver	g %*	$91.0 \\ 5.23$	$65.2 \\ 3.18$	$66.6 \\ 3.17$	$71.1 \\ 2.29$	$83.3 \\ 2.58$	3.29	0.78
Spleen	g % *	$0.81 \\ 0.047$	$\begin{array}{c} \textbf{1.24} \\ \textbf{0.060} \end{array}$	$\frac{1.64}{0.078}$	$0.92 \\ 0.030$	$0.72 \\ 0.022$	0.047	0.017
Adrenals	g %*	$\frac{0.27}{0.016}$	$\frac{0.14}{0.007}$	$0.21 \\ 0.010$	$0.23 \\ 0.007$	$0.25 \\ 0.008$	0.010	0.003
Marrow	g %*	$\frac{48.3}{2.78}$	$\frac{37.3}{1.82}$	$\begin{array}{c} \textbf{48.3} \\ \textbf{2.30} \end{array}$	$62.4 \\ 2.01$	$61.0 \\ 1.89$	2.16	0.30
Bone	g %*	$\frac{67.8}{3.90}$	$75.8 \\ 3.69$	85.6 4. 07	$109.9 \\ 3.54$	$103.5 \\ 3.22$	3.78	0.36
"Whole bone"	g %* .	$116.2 \\ 6.68$	113.1 5.51	$133.9 \\ 6.37$	$172.4 \\ 5.55$	$164.4 \\ 5.11$	5.84	0.54
Whole bone-ash	g %*	$\frac{36.3}{2.08}$	$\frac{43.1}{2.10}$	$\begin{array}{c} 46.7 \\ 2.23 \end{array}$	$\frac{63.7}{2.06}$	$\frac{60.7}{1.88}$	2.07	0.08

^{* %} of body wt.

values are in agreement with those found by Nye.¹ Comparing it with other organs, the bone marrow is roughly equal to two-thirds the weight of the liver (53 to 87%), and 50 times (30 to 86) the weight of the spleen, and 250 times (174 to 288) the weight of the adrenals. Correlation with the body weight is unsatisfactory because of the individual differences in body fat content. As an example, Rabbits No. 4 and 5 (Table II) had large fat depots.

Although individual animals showed differences in the total bone marrow, the marrow for each of the bones was distributed in constant proportions to the total. Almost half of the bone marrow was found in the hind legs (including the ossa coxæ). The head contained the smallest amount in proportion to its bone content. The variation in the distribution in the vertebræ was slightly accentuated since one rabbit had only 6 cervical and another only 6 lumbar vertebræ and the number of caudal vertebræ varied from 15-18

The distribution of bone and bone-ash differed from that of the marrow, chiefly because of the relatively large amount of inorganic matter in the skull and mandibles.

From the values given in Table I, the amounts of active and inactive marrow can be estimated. The actual amounts depend upon the age and condition of the animal, and as to whether the partly active marrow is considered active or inactive. If the marrow of the pedes, manus, and caudal vertebræ is classified as inactive, 85-90% of the total marrow is active. Nye¹ estimated the active bone marrow in rabbits as 75%.

Conclusions. A study of distribution of bone marrow in rabbits showed 47% of the marrow in the hind legs, 18% in the fore legs, 6% in the ribs, sternum, and clavicles, 9% in the head, and 20% in the vertebræ. The distribution of bone marrow, bone and boneash in the individual bones is recorded in the tables.