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Received October 13, 1955. P.S.E.B.M., 1956, v91.

Residual Organ Blood Volume of Cattle, Sheep and Swine.* (22160)

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In quantitative studies involving investigations of tissue distribution of blood-borne substances a correction for the residual organ blood may be essential. This is of special importance in short time radioisotope distribution experiments where retained tissue blood may be sufficient to contribute significantly to the total activity recovered. Specific studies based upon whole tissue extractions measure not only the substance in the cells of the tissues, but also that existing in the blood which is trapped within these tissues at the time of sampling. Therefore, in tracing the distribution of an ingested or injected substance by extraction from various organs, it is necessary to know the proportion of the material that is from the tissue and that which is from the blood itself. When the relative proportions of blood and tissue are known, precise quantitative studies on movement and tissue composition may be used to substantiate microscopic examinations and organ weight changes. The total organ weights for farm animals have been reported by several investigators(1-3), and procedures and values for blood volume of cattle, sheep and swine are in the literature(4-6). Lewis *et al.*(7) have reviewed procedures and reported the total or-

gan blood volume measurements for the rat. Other investigators have made *in vitro* and *in vivo* measurements of total organ blood volumes in laboratory animals(8,9) but there were no data available from the literature directly related to the residual blood present in organs and tissues of farm animals slaughtered by the conventional procedures.

This report is concerned with the residual blood values as related to age and species for the various organs and tissues of farm animals routinely sacrificed and bled out during current nutritional investigations.

Material and methods. The cattle, sheep and swine used in these studies were normal, healthy individual animals maintained under general farm management conditions. Total red blood cell volumes were obtained by the injection of P^{32} labeled homologous erythrocytes as previously described(4). The organ blood values were procured as follows: (a) blood samples were drawn from the animal; (b) whole blood was incubated with P^{32} ; (c) red blood cells were washed sufficiently to remove any radioactivity not in the cells and (d) the cells were reconstituted to the original volume with the animal's own plasma. (e) A known volume of suspended labeled cells was re-injected into the immobilized animal(10) and after mixing had occurred, blood samples were taken from the jugular vein of the animal, usually at 5 and at 15 minute intervals for radioactivity measurement and blood volume determinations. (f) The animal was then sacrificed by a blow

* This article is published with approval of the Director of University of Tennessee Agricultural Experiment Station. The technical assistance of G. W. Royster and T. G. Clark is gratefully acknowledged. The radiophosphorus was procured from the Oak Ridge National Laboratory on allocation from the U. S. Atomic Energy Commission.

TABLE I. Mean Organ Weights for Cattle, Swine and Sheep.

Species	Age (mo)	No. of animals	Mean wt (lb)	Fresh wt as % of body wt $\pm$ stand. dev.					
				Blood*	Liver	Kidney†	Spleen	Lungs	Heart
Cattle	.5-3	12	88	8.4	2.31 $\pm$.71	.230 $\pm$.035	.359 $\pm$.106	1.57 $\pm$.42	.767 $\pm$.242
	6	15	384	6.0	2.24 $\pm$.19	.124 $\pm$.024	.266 $\pm$.021	.821 $\pm$.12	.457 $\pm$.087
	Aged	8	923	5.6	2.17 $\pm$.06	.130 $\pm$.032	.256 $\pm$.024	.859 $\pm$.120	.471 $\pm$.051
Sheep	.5	5	10	9.3	1.69 $\pm$.24	.560 $\pm$.30	.320 $\pm$.28	1.65 $\pm$.05	.740 $\pm$.01
	4	6	80	6.5	1.84 $\pm$.24	.140 $\pm$.02	.240 $\pm$.03	1.08 $\pm$.10	.360 $\pm$.12
	Yearlings	8	118	5.9	1.30 $\pm$.29	.120 $\pm$.02	.260 $\pm$.02	1.00 $\pm$.30	.390 $\pm$.08
Swine	.5	7	10	7.2	2.52 $\pm$.25	.415 $\pm$.046	.297 $\pm$.125	1.29 $\pm$.22	.609 $\pm$.163
	2	11	35	6.7	2.10 $\pm$.26	.299 $\pm$.048	.220 $\pm$.066	1.21 $\pm$.22	.472 $\pm$.091
	5	6	102	6.3	2.66 $\pm$.34	.242 $\pm$.061	.232 $\pm$.051	.95 $\pm$.20	.403 $\pm$.101
	Aged	6	654	3.5	1.859 $\pm$.22	.820 $\pm$.0228	.218 $\pm$.029	.834 $\pm$.117	.319 $\pm$.039

* ml of blood/100 g body wt.

† Mean wt of one kidney only.

on the head, hoisted by the rear legs and bled out for 5 minutes. The organs and tissues were immediately excised, blotted free of excess blood and weighed, (g) whole organs or 10 to 15 samples were taken from each organ or tissue being studied for the measurement of radioactivity, and (h) these average values were used for the estimation of the residual blood present. All tissue samples were weighed and wet ashed with nitric acid, made to volume and aliquots taken for radioactivity measurements by conventional procedures as described by Hansard *et al.*(4). The milliliters of blood per 100 g of tissue were then calculated from the isotope concentration in the samples and the blood volume standard (5).

Results. Organ weights of farm animals. The mean unit weights of the various organs of cattle, sheep and swine employed are listed in Table I as a percentage of the live body

weight. These average values compare favorably with those reported by other investigators(1-3). It is noted that there was more difference due to weight of animal than to species. The very young of all species showed a higher blood volume and slightly higher mean organ weights. These values are included to provide a means of estimating the total residual blood present in whole organs and tissues.

Residual blood in cattle organs and tissue. The residual blood in organs of 7 cattle in 2 age groups is shown in Table II. There was a wide variation between individual tissues but the total blood volume values were similar to those reported previously(4). In these animals the spleen retained the highest concentration of residual blood, followed by the lungs, liver, kidney, pituitary, adrenal, heart, pancreas and muscle in that order. Age appeared to affect concentration of retained

TABLE II. Residual Organ Blood Volume of Cattle at Various Weights.

Wt (lb)	Total blood vol (ml/100 g)	ml blood/100 g (fresh wt)								
		Spleen	Lungs	Liver	Kidney	Pituitary	Adrenal	Heart	Pancreas	G. muscle
70	7.6	31.6	23.2	10.4	3.9	2.8	3.1	2.9	2.3	1.1
78	9.1	29.7	26.2	6.1	3.4	4.0	2.8	2.2	2.2	.8
83	8.5	32.6	25.8	7.2	3.7	3.4	3.2	2.7	3.1	1.0
Avg 77	8.4	31.3	25.1	7.9	3.7	3.4	3.0	2.6	2.5	.96
$\pm 7.7^*$	$\pm .9$	± 1.7	± 1.8	± 2.5	$\pm .3$	$\pm .7$	$\pm .2$	$\pm .4$	$\pm .5$	$\pm .12$
800	5.6	22.1	23.4	9.5	7.0	4.6	3.9	4.4	4.5	.7
758	5.8	39.3	18.2	6.0	6.9	4.3	3.5	3.0	3.2	.9
834	5.7	39.4	18.2	7.3	5.9	3.7	3.8	3.4	2.8	1.2
992	5.4	37.0	21.5	8.0	6.6	5.8	4.1	3.9	2.9	1.3
Avg 846	5.6	34.5	20.3	7.7	6.6	4.6	3.8	3.7	3.3	1.0
± 115	$\pm 2.$	± 3.6	± 2.5	± 1.7	$\pm .5$	$\pm 1.$	$\pm .3$	$\pm .7$	$\pm .8$	$\pm .2$

* Mean $\pm$ S.D.

TABLE III. Residual Organ Blood Volume of Sheep at Various Weights.

Wt (lb)	Total blood vol (ml/100 g)	ml blood/100 g (fresh wt)									
		Spleen	Lungs	Liver	Kidney	Heart	Pan- creas	Adrenal	Pitui- tary	Loin muscle	G. muscle
6	10.0	33.0	16.3	10.3	6.2	5.3	4.0	1.7	1.9	2.6	2.0
10	9.3	36.1	18.2	9.8	5.7	6.3	3.2	1.9	2.2	1.3	1.4
12	8.5	35.4	20.1	8.6	4.8	6.2	2.1	2.1	2.0	1.8	1.6
Avg 9	9.3	34.8	18.2	9.6	5.6	5.9	3.1	1.9	2.0	1.9	1.7
± 3.5*	± .9	± 1.8	± 2.2	± 1.	± .8	± .6	± 1.1	± .2	± .2	± .8	± .4
96	6.2	33.0	29.4	9.1	5.3	5.5	4.0	2.2	2.1	1.1	1.5
102	6.1	36.8	29.0	6.6	5.3	4.4	2.5	4.2	2.0	.9	.7
136	5.5	34.2	21.5	8.6	6.4	5.5	3.3	3.1	2.5	1.5	1.2
Avg 111	5.9	34.7	26.6	8.1	5.4	5.1	3.3	3.2	2.2	1.2	1.1
± 24	± .4	± 2.2	± 4.7	± 1.5	± .6	± .6	± .9	± 1.2	± .3	± .4	± .5

* Mean ± S.D.

blood, but the differences between old and young cattle were not consistent nor sufficient for significance in this study.

Residual blood in sheep organs and tissue. The milliliters of blood per 100 g of fresh tissue are shown for old and young sheep in Table III. The trend followed essentially the same pattern as was indicated for cattle, and the milliliter percentage of residual blood closely paralleled that for organs and tissues of cattle. When consideration is given to the fact that one-third and one-fourth of the weight of the spleen and of the lungs respectively were due to blood itself, the significance of this dilution factor for the interpretation of tissue analysis values can be realized. Other tissue and organ blood values ranged from one to 10% on a fresh weight basis.

Residual blood in swine organs and tissue. Residual blood determinations were made in tissues and organs from 10 swine at different ages and weights as indicated in Table IV. The calculated milliliters of residual blood per 100 g of fresh tissue show the lungs to be highest, followed by the spleen, kidney, liver, adrenals, heart, pancreas, loin muscle and gastrocnemius muscle in that order. There appeared to be a general tendency toward a higher concentration of residual blood in organs and muscle of the younger animals.

Discussion. The isotope dilution procedure using P^{32} labeled red cells has been found convenient and reliable for the quantitative measurement of the residual blood in organs and tissues of farm animals. In radioisotope investigations and specific kinetic

studies with blood-borne substances these values require emphasis. Because of the intimate relationship between the cells of the body and the blood, inherent errors in analysis may be encountered due to residual blood trapped in the tissues at the time of slaughter. For the most part, routine sampling is made from animals that have been bled out; therefore, total organ blood volume(6-8) was not considered a necessity in this study. It is pointed out that since the residual blood calculations are based upon total blood volume values, consideration should be given to some of the more important sources of error when the isotope dilution procedure is employed. These errors invariably leading to a high blood volume value include: a) loss of label from cells injected, b) non-quantitative injection of cells into the blood stream and, c) removal of label from circulating blood following administration. Every precaution was taken in this study to avoid or to correct these errors, and by management practices to avoid physiological changes known to affect the normal animal. Since whole organs could not always be analyzed conveniently, multiple samples were taken routinely from similar areas of the various organs and processed immediately. The percent of the total blood volume for individual organs showed considerable variation, but residual blood per gram within tissues was remarkably constant. There was a general tendency for a lower residual blood concentration in tissues and organs of older and heavier animals, and a decrease for muscle tissue with increased age

TABLE IV. Residual Organ Blood Volume of Swine at Various Weights.

Wt (lb)	Total blood vol (ml/100 g)	ml blood/100 g (fresh wt)								
		Lungs	Spleen	Kidney	Liver	Adrenal	Heart	Pancreas	Loin muscle	G. muscle
8	7.1	26.2	13.1	6.5	8.1	5.0	4.0	2.2	2.2	1.0
7	7.4	27.8	12.3	6.3	7.8	6.0	4.6	2.1	2.2	1.2
10	7.0	25.2	12.5	6.8	8.3	5.7	3.3	1.7	2.2	1.0
12	6.9	24.8	12.2	5.3	6.4	6.3	3.1	2.5	2.1	.9
Avg 9	7.1	26.0	12.5	6.0	7.7	5.8	3.8	2.1	2.2	1.0
± 2*	± .2	± 1.5	± .4	± .7	± .9	± .6	± .7	± .4	± .0	± .1
28	6.8	22.9	15.1	7.1	5.1	5.4	3.4	2.1	1.4	.79
32	6.5	28.2	16.3	7.5	5.9	5.4	3.1	2.3	1.9	.80
78	6.4	23.4	12.5	6.9	6.1	4.0	2.8	2.3	1.2	.82
Avg 46	6.6	25.0	14.6	7.2	5.7	5.0	3.1	2.2	1.5	.80
± 30	± .2	± 3.1	± 2.2	± .4	± .6	± .8	± .4	± .1	± .4	± .02
644	3.3	23.1	9.9	5.4	8.4	3.6	3.2	1.9	.92	.89
668	3.1	24.8	10.0	11.1	7.3	5.4	4.1	2.5	.90	.73
547	4.0	24.1	10.1	10.7	8.2	4.4	3.5	2.4	.92	.82
Avg 620	3.5	24.0	10.0	9.1	7.9	4.5	3.6	2.3	.91	.84
± 71	± .5	± 1.	± .1	± 3.4	± .6	± 1.1	± .5	± .4	± .01	± .08

* Mean ± S.D.

and weight. Little difference between species was noted except that the spleen, liver, pancreas and gastrocnemius muscle of cattle and sheep retained more blood than did those of swine. The lower concentration of blood in muscle noted especially in aged swine may well have been due to the increased fat content of these animals.

Summary. The residual blood in organs and tissue, as a function of age and weight, for cattle, sheep and swine sacrificed by conventional methods was determined by the isotope dilution procedure using P^{32} labeled homologous erythrocytes. Organ weights and total blood volume compared favorably with literature reports. Neither age, weight nor species appeared to influence significantly the ml of tissue residual blood/100 g fresh weight, except that the spleen, liver, pancreas, and gastrocnemius muscle of cattle and sheep retained more blood than did those tissues of swine. There was a general tendency for higher residual blood concentrations in organs and tissue of younger animals, but these findings were not consistent. Adrenals, heart, pancreas, kidney, liver, pituitary and muscle tissue retained less than ten ml of blood/100 g fresh weight, while lungs and spleen re-

tained more residual blood. It is suggested that these values may provide a basis for estimated corrections in quantitative investigations for distribution of blood-borne substances in farm animals.

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Received October 13, 1955. P.S.E.B.M., 1956, v91.