702736

REPOSITORY REECo
COLLECTION Tulane University Journals
BOX No. N/A
FOLDER N/A

Tissue distribution, kinetics, and biologic half-life of Mg^{28} in the dog[1]

R. LAZZARA, K. HYATT, W. D. LOVE, J. CRONVICH, AND G. E. BURCH

Department of Medicine, Tulane University School of Medicine, and Charity Hospital of Louisiana, New Orleans, Louisiana

LAZZARA, R., K. HYATT, W. D. LOVE, J. CRONVICH, AND G. E. BURCH. *Tissue distribution, kinetics, and biologic half-life of Mg^{28} in the dog.* Am. J. Physiol. 204(6): 1086–1094. 1963.—Mg^{28} of a high specific activity was used in these studies. It was rapidly injected intravenously into 12 dogs and the concentration-time course curves in plasma were obtained. Urinary and fecal excretion was followed in seven animals. The dogs were killed 7 min to 68 hr following injection. Seventy-four tissues were sampled and assayed for Mg^{28} concentration. Plasma Mg^{24} was determined by flame spectrophotometry. With these data it was possible to follow the concentration-time course of Mg^{28} in the various tissues as well as to calculate their "exchanging" Mg^{24} content and effective rates of exchange. A variation in the pattern of uptake and in concentration of exchanging Mg^{24} was noted among the different tissues. During the interval from 24–68 hr, body exchanging Mg^{24} space and mass were found to range from 3.2–3.8 liters/kg and 4.9–5.7 mEq/kg, respectively. The biologic half-life of Mg^{28} in the dog estimated from excretion data was 11 days.

MAGNESIUM IS PRIMARILY an intracellular ion with important functions in cellular metabolism. The availability of the radioisotope Mg^{28} for research led to renewed interest in magnesium, a trend reflected by the number of recent reviews of magnesium metabolism (1–4). A number of studies utilizing Mg^{28} have been published concerning aspects of normal magnesium metabolism in various mammalian species. Several reports deal primarily with the kinetics and distribution of Mg^{28} in tissues of a few animal species (4–11). Mg^{28} first became available only in low specific activity. Because of the low specific activity, the short half-life (21.3 hr), and the high cost of the isotope, investigations were discouraged. The use of sufficient quantities of isotope to allow prolonged sampling of tissues necessitated the administration of metabolically significant and toxic quantities of carrier magnesium which interfered with the conduction of satisfactory studies of normal metabolic phenomena. In February 1961, Mg^{28} of higher specific activity was made available by the Brookhaven National Laboratories so that the latter difficulty could be largely circumvented. The present experiments conducted with this Mg^{28} of high specific activity were undertaken to study the rate of plasma clearance and excretion of Mg^{28} and the kinetics of its distribution in the tissues of normal dogs. In general the plan of the studies was similar to those reported previously from this laboratory for other radioelements (12, 13).

Received for publication 23 July 1962.

[1] Supported by grants from the Public Health Service, the Rowell A. Billups Fund for Research in Heart Disease, and the Rudolph Matas Memorial Fund for the Kate Prewitt Hess Laboratory.

MATERIAL AND METHODS

Thirteen healthy mongrel dogs were carefully maintained in the laboratory kennel under metabolically controlled conditions for a minimum of 30 days prior to their use in these experiments. Their diet consisted of Purina dog chow with occasional supplementation with Pard (Armour). Mg^{28} in the form of $MgCl_2$ received from Brookhaven National Laboratories was neutralized with sodium hydroxide and filtered. The specific activity varied from 10.8 μc/mg to 25.3 μc/mg of $MgCl_2$ at the time of intravenous injection. The usual dose administered intravenously was 180 μc, the amount of carrier magnesium varying from 6.9 mg to 23.5 mg. The dogs were anesthetized with intravenous pentobarbital. Indwelling needles were placed in a femoral vein and a femoral artery. In six animals heparinized arterial samples were obtained in rapid succession for the first 2.5 hr, at intervals from a few seconds to a few minutes. Samples of femoral venous blood were then drawn twice daily for long-term follow-up studies. A sample of arterial blood was obtained immediately before death in all animals. Plasma was separated from the erythrocytes in all samples. Complete excretion data were obtained for Mg^{24} in seven dogs.

The dogs were sacrificed at intervals varying from 7 min to 68 hr following administration of the isotope by rapidly opening the chest and clamping first the inflow and then the outflow vessels of the heart. Immediately following death the major organs were removed, drained of blood, and weighed. Samples of these organs and other tissues were collected, blotted with a dry cotton gauze sponge, and weighed. Seventy-four different tissues were sampled for study.

0024172

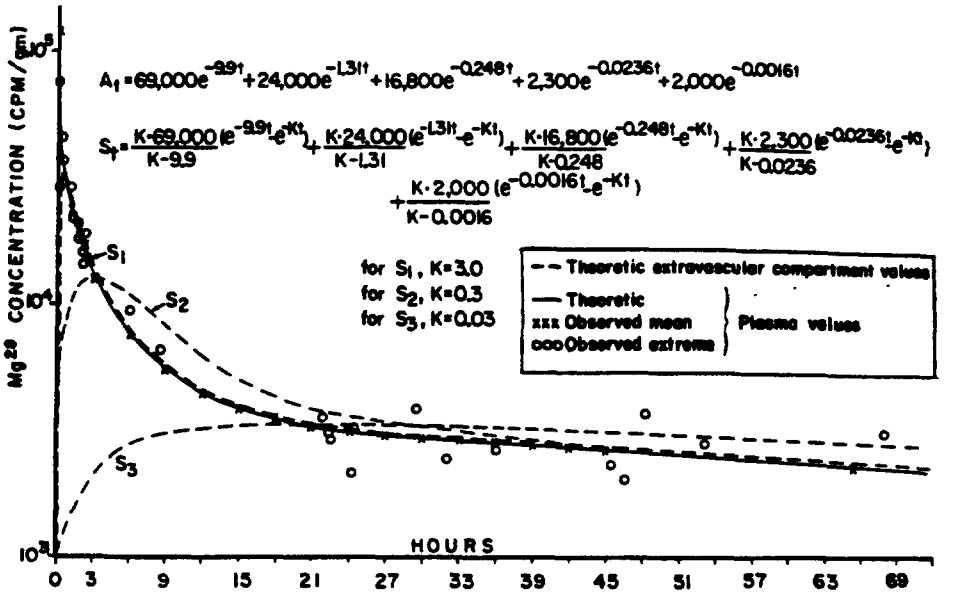

FIG. 1. Concentration-time course of Mg^{28} observed for plasma and theoretic curves for plasma and extravascular compartments. The crosses and circles represent the mean and extreme values, respectively, observed for the plasma. The solid line represents the calculated curve for the plasma whereas the interrupted line represents the calculated curves for three extravascular compartments with different rates of exchange. The curves for the theoretic compartments and the theoretic plasma curve are time courses of specific activity whereas that observed for the plasma is a time course of concentration of tracer only. The units for the ordinate are shown only for concentration of the tracer in order to present the magnitude of the values observed in the plasma.

A litter of nine embryos (approximately 40–50 days gestation) was present in the one dog sacrificed at 68 hr. This offered an opportunity to sample the tissues of the embryos for comparison with those of the mother. From each embryo selected organs (heart, lungs, kidneys, brain, liver, and spleen) were removed and analyzed and the remaining carcasses were also analyzed in entirety.

The samples of tissue were digested in hot nitric acid. When digestion was complete, the excess nitrates were driven off by adding formic acid according to the method of Sheppard and Martin (14). The resulting solution was diluted with distilled water to a total volume of approximately 5 ml.

All samples including plasma were analyzed for Mg^{28} gamma emission with a Baird-Atomic model 81 OB well-type sodium iodide crystal counter. Plasma Mg^{24} was determined with a Zeiss PMQ II flame spectrophotometer by I. MacIntyre. The method utilized was described previously by him (15).

Considerations of method for analysis of data. In the analysis, all calculations of radioactivity were reduced to a dose of 10,000,000 counts/min kg of body wt. The theoretic and mathematic approach considered the blood plasma as the central compartment of a centrally exchanging mammillary system. All the tissues were treated, respectively, as single homogeneous compartments communicating directly with the plasma and extracellular fluid. This oversimplification was necessary in this study since no differentiation was possible between various cell types or between intracellular magnesium and that in interstitial fluid. In this report the term "exchanging" magnesium refers to the tissue magnesium actively exchanging with the plasma at any time, t. It does not necessarily indicate all body magnesium was involved in the exchange. It is necessary to differentiate between the "exchanging" (time-dependent) and the "exchangeable" compartment. Because of the lack of a sufficiently accurate method, Mg^{24} was not measured in the tissues.

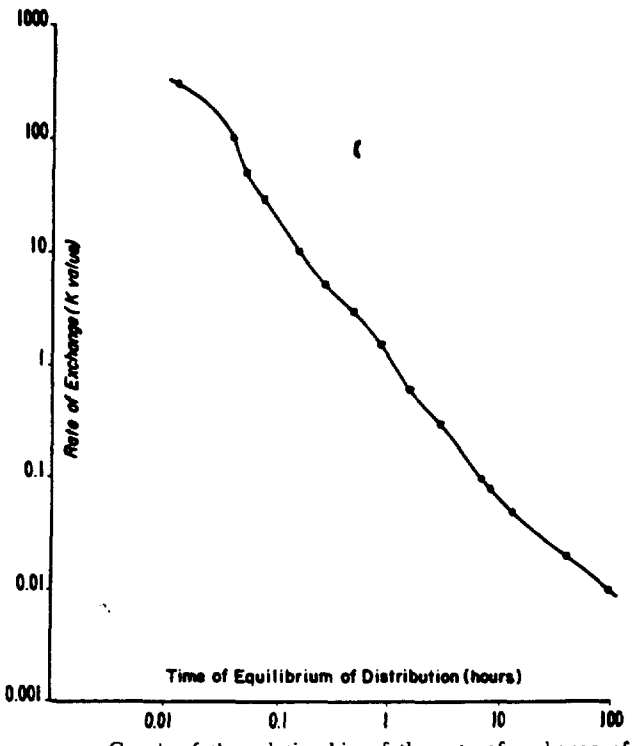

FIG. 2. Graph of the relationship of the rate of exchange of Mg^{28} for an extraplasmal compartment to the time at which equilibrium of distribution would be expected to be reached between that compartment and the plasmal compartment. This curve obtained from the theoretic concentrations noted in the text makes it possible to estimate readily when equilibrium of distribution would be expected for various rates of exchange between the plasma and an exchanging compartment.

Therefore, it was not possible to determine tissue specific activity for comparison with plasma specific activity. This relationship provides the most accurate means of determining the time at which equilibrium of distribution of any tissue with plasma is achieved. However, following single intravenous administration of a tracer

0024173

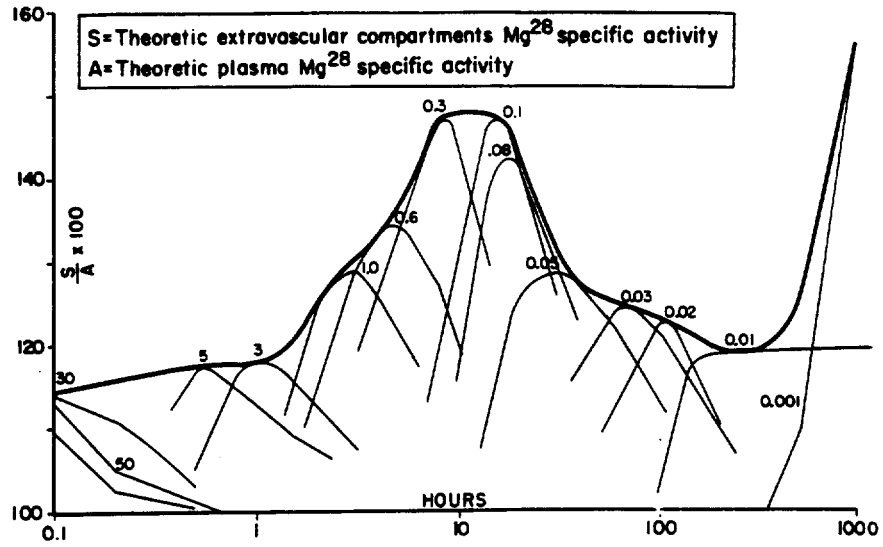

FIG. 3. Effect of lag phenomenon on tissue Mg^{28} specific activity. The heavy solid line connects the peaks of the smaller curves and represents the maximum lag effect produced at any given time for organs with various rates of exchange. The individual curves were constructed from the formula shown on the ordinate using a different rate of exchange (k value shown by small numbers) for each curve. Thus, the value above 100 represents the per cent error in calculation due to the effects of lag.

when a tissue which communicates only with the central compartment reaches equilibrium of distribution with the plasma, its Mg^{28} concentration ceases to rise. Therefore, examination of the concentration-time course of Mg^{28} of the various tissues allows one to estimate fairly accurately the time at which equilibrium was established (12). The concentration-time course curves were not ideal because of the relatively small number of observations and the variation among animals. Therefore, the time of peak concentration of radioactivity selected in these experiments represents only an approximation of the time at which equilibrium of distribution was achieved. Nevertheless, at the moment that equilibrium of distribution is established, the size of the compartment of exchanging magnesium in the tissue can be calculated by the following formula:

$$\text{exchanging tissue } Mg^{24} = \frac{\text{tissue } Mg^{28}(\text{counts/min g}) \times \text{plasma } Mg^{24}(\text{mEq/kg})}{\text{plasma } Mg^{28}(\text{counts/min g})} \quad (1)$$

The above equation applies only at the time the specific activity of the tissue compartment first becomes equal to that of the plasma compartment, i.e., the time at which the concentration of Mg^{28} in the tissue compartment reaches its peak. Following this time the concentration of Mg^{28} and the specific activity of Mg^{28} in the tissue will decline but the decline in specific activity of the tissue may "lag" behind the decline in specific activity of the plasma. This will occur if the rate of exchange of the tissue magnesium with plasma is slower than the rate of decline of the specific activity of the plasma. This has been termed the lag phenomenon, which has been discussed in detail in a previous communication from this laboratory (12). Application of the above equation during the period after the peak of specific activity has been reached in a slowly exchanging compartment leads to an overestimation of magnitude of the exchanging Mg^{24}. It is possible to estimate this error. Basically, the analysis requires the expression of the time course of plasma radioactivity (A_t) (Fig. 1) which is as follows:

$$A_t = A_{01}e^{-b_1t} + A_{02}e^{-b_2t} \cdots + A_{0n}e^{-b_nt} \quad (2)$$

where t is elapsed time in hours after injection of the isotope, A_{01}, $A_{02} \cdots A_{0n}$ are plasma radioactivity at t and $b_1, b_2 \cdots b_n$ are fractional rates of decline of plasma radioactivity. The values for b are determined by the formula:

$$b = \frac{\log e^2}{T_{1/2}} \quad (3)$$

where $T_{1/2}$ refers to the half-time of the linear exponential.

The tissue radioactivity, S, is calculated by substituting the values for fractional rates of turnover (k) of tissue magnesium in the formula:

$$S_t = \frac{kA_{01}}{k - b_1}(e^{-b_1t} - e^{-kt}) + \frac{kA_{02}}{k - b_2}(e^{-b_2t} - e^{-kt}) \cdots + \frac{kA_{0n}}{k - b_n}(e^{-b_nt} - e^{-kt}) \quad (4)$$

where S_t is tissue radioactivity at time t.

If one expresses the radioactivity in terms of specific activity, S_t is obtained directly from the above equation. However, in order to obtain concentration of tracer one must multiply S_t by a factor (the ratio of tissue concentration of stable isotope to plasma concentration).

Theoretical radioactivity-time course curves for various compartments can be constructed (Fig. 1) by substituting various values of k in the formula. By determining the times of intersection of these curves with the calculated plasma curves plotted in terms of specific activity it is possible to construct a graph showing the theoretical times at which equilibrium of distribution (or maximal concentration of tracer) is reached in tissues with various theoretic rates of turnover (Fig. 2). The

0024174

TABLE 1. *Plasma Mg^{24} at time of sacrifice**

Dog No.	Hr	Min	Concn. Mg^{24}
1	1	33	2.08
2	15	44	
3	24	30	2.16
4	68	8	1.42
5	4	3	1.36
6	9	20	1.42
7		19	1.90
8	22	23	1.37
9	46	20	1.50
10	2	30	1.42
11		7	1.29
12	36	5	1.42
13	56	57	1.37

* Conversion from mEq/liters to mEq/kg was based on the assumption of a plasma specific gravity of 1.053. The measurements were determined by I. MacIntyre.

approximate value for the actual rate of turnover in sampled tissues can be estimated by matching the experimentally recorded time of maximal concentration of tracer with the abscissa of the graph and obtaining the rate of exchange from the ordinate.

The ratio of S_t to A_t after equilibrium has been reached will exceed unity (or 100%) for a certain time interval which will depend on the rate of exchange of the particular compartment (see above). By plotting this ratio for the time intervals at which it exceeds unity for various theoretic compartments, one may obtain a series of partial time-course curves for a corresponding set of theoretic compartments (Fig. 3). The peaks of this series of curves (connected by the heavy solid line) will delimit the maximum error which can be produced by lag for any compartment at any time, t.

From the balance data, the plasma-equivalent space and exchanging mass of Mg^{24} as functions of time (16) and the biologic half-life of Mg^{28} in the dog (17) were determined.

RESULTS

The concentration-time course of plasma radioactivity is shown in Fig. 1. This curve was analyzed graphically into five linear exponential components. The equation for the curve is shown in Fig. 1. The concentration of Mg^{24} in the plasma of each dog just prior to sacrifice appears in Table 1.

Concentration-time course curves of Mg^{28} for some of the major organs appear in Fig. 4. Each value on each curve was obtained for a different dog at the time of sacrifice. The highest concentrations occurred in the heart and the lowest in skeletal muscle (Fig. 4). The organs had different rates of uptake of the tracer. Although such curves are available for about 74 different tissues, they could not be charted satisfactorily in a single figure. Table 2 does provide data for all the sampled tissues.

The fraction of the doses administered which accumulated in some of the major organs at each time interval is summarized by Fig. 5. Among the values shown in Fig. 5 the liver accumulated the largest fraction of the total administered dose although the liver did not

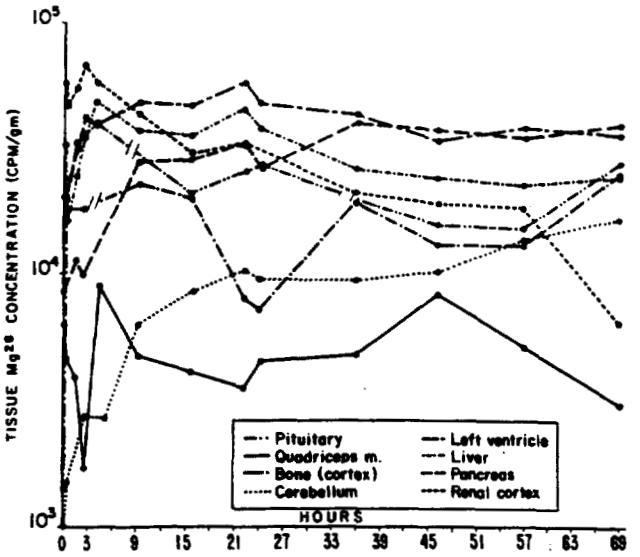

FIG. 4. Concentration-time course of Mg^{28} for various tissues.

develop the highest concentration of isotope. These curves show the different rates of uptake of Mg^{28}. In many of the curves there was an early peak, probably reflecting high concentration in the blood and extracellular fluid contained in the tissues, followed by a secondary peak, reflecting the parenchymal uptake.

Table 2 lists all the tissues studied with the corresponding times at which equilibrium of distribution was established, the k values, and the magnitude of exchanging Mg^{24}. Chemically determined Mg^{24} concentrations obtained from the literature for some canine organs are listed for comparison (Table 2). The range of times at which maximal concentrations were reached is also tabulated. Determination of the absolute peak of the concentration-time course curve was not possible. Likewise, a range of k is also shown for most of the tissues. When two distinct peaks of Mg^{28} concentration were found both values are shown. To minimize the error produced by the lag phenomenon (Fig. 3), the amount of exchanging Mg^{24} for each tissue was calculated for the three longest periods of time after administration of the tracer (46, 57, and 68 hr) (Table 2). Although the error produced by lag in calculating the quantity of exchanging Mg^{24} is least at the times selected, there may be an overestimation of the magnitude of exchanging Mg^{24} by not more than 25% and probably less.

The average calculated exchanging Mg^{24} for some organs and remaining carcasses of the nine embryos and for the mother sacrificed at 68 hr are shown (Table 3).

The cumulative data on urinary and fecal excretion of the administered Mg^{28} shown in Table 4 were compiled from observations on seven dogs. Three to twenty per cent of the injected Mg^{28} was excreted in the urine and less than 3% in the feces during the first 24 hr. Maximal amount excreted in the feces was 4.5% in one dog by 46 hr after administration of the tracer. At the end of the longest period of observation urinary excretion accounted for less than 21% of the total ad-

0024175

TABLE 2. *Master table*

Organ	Time of Maximum Count, hr		Estimated k, Fractional Rate of Exchange/hr		Estimated Exchanging Mg^{24} Content, mEq/kg			Chemically Determined Organ Mg^{24} Content, mEq/kg	Ref.
	Peak	Range	k	Range	46 hr, 20 min	56 hr, 57 min	68 hr, 8 min		
Heart									
Left ventricle	22.4	9.3–24.3	.032	.069–.030	25.3	22.8	23.1	17.35–19.83	(18)
Right ventricle	22.4	9.3–24.3	.032	.069–.030	24.9	23.2	23.1	17.35–19.83	(18)
Epicardial myocardium	22.4	9.3–24.3	.032	.069–.030	25.1		25.4		
Papillary muscle	22.4	9.3–24.3	.032	.069–.030	25.5	22.8	22.0		
Intervent. septum	22.4	9.3–22.4	.032	.069–.032	24.9	22.8			
Left atrium	22.4	9.3–22.4	.032	.069–.032	17.6	16.1	12.7		
Right atrium	22.4	9.3–22.4	.032	.069–.032	16.3	13.7	12.7		
Valve	0.32/36.1		4.2/.022		8.7	7.8			
Pericardium	1.5/36.1*		.64/.022		4.0	3.7	3.6		
Pericardial fluid†						1.12			
Vessels									
Thoracic aorta	0.12/22.4*		14.0/.032		8.3	7.5	7.1		
Abdominal aorta	0.12/57*		14.0/.015		5.7	9.3	7.5		
Pulmonary artery	0.32/22.4*		4.2/.032		8.0	7.5			
Pulmonary vein	0.32/22.57*		4.2/.032–.015		7.8	18.9			
Sup. vena cava	0.32/24.33*		4.2/.030		9.6	9.0	10.0		
Inf. vena cava	0.32/22.38*		4.2/.032		2.6	5.1	3.6		
Portal vein	0.12/57*		14.0/.015		7.5	1.7	9.5		
Lung	9	9.0–36.1	.069–.022	.069–.022	11.4	11.4	11.9		
Trachea	4	2.5–4.0	.36/3.4	.36/3.4	3.4	10.8	10.9		
Pleura	0.32/48.3*		4.2/.0175		6.8	5.0	4.5		
Digestive system									
Salivary gland	22.4	4.0–68.1	.032	.20–.013	22.6	16.2	20.3		
Esophagus	0.32/24.3*		4.2/.030		7.5	7.1	7.47		
Stomach	15.7	9.3–36.0	.042	.069–.022	21.1	13.1	47.8		
Stomach contents	2.5		.36		3.4	3.1	3.0		
Small intestines	9.3	2.5–22.4	.069	.36–.032	15.0	17.0	17.7		
Small intestine contents	2.5		.36			10.8	11.8		
Large intestines	2.5	2.5–24.3	.36	.36–.030	13.3	13.0	14.8		
Large intestine contents	36.1	22.4–48.3	.022	.032–.0175	14.4	10.8	10.0		
Liver	4.0	2.5–24.3	.20	.36–.030	17.6	13.7	15.7	13.5–14.59	(19, 20)
Gall bladder	0.12/15.7*		14.0/.042		10.4	6.6	9.0		
Bile	4.0	4.0–36.0	.20	.20–.022	15.7	7.5	8.4		
Pancreas	36.1	22.4–68.1	.022	.032–.013	27.3	21.3	25.0		
Peritoneum	0.32/68*		4.2/.013		4.1	5.2	13.3		
Urogenital system									
Renal cortex	2.5	0.12–4	.20	14.0–.20	13.6	11.1	4.1		
Renal medulla	2.5	0.12–4	.20	14.0–.20	7.9	5.1	7.1		
Ureter	15.7	1.5–24.3	.042	.64–.030	8.7	10.1	6.8		
Bladder	15.7	15.7–57.0	.042	.042–.015	11.8	11.6	10.7		
Uterus	36.1	4.0–68.1	.022	.36–.013	9.0		8.0		
Ovary		1.5–48.3		.64–.0175	8.8		10.0		
Testicle†						12.1			
Spermatic cord†						11.5			
Epididymis†						9.3			
Prostate†						10.0			
Endocrine system									
Pituitary	4.0	1.5–4.0	.20	.64–.20	11.4	9.2	16.4		
Adrenal	2.5/24.3*	1.5	.20/.030	.64	12.5	14.4	14.9		
Lymph node	15.7	9.3–46.3	.042		17.1		9.9		

0024176

TABLE 2. *Continued*

Organ	Time of Maximum Count, hr		Estimated k, Fractional Rate of Exchange, hr		Estimated Exchanging Mg^{24} Content, mEq/kg			Chemically Determined Organ Mg^{24} Content, mEq/kg	Ref.
	Peak	Range	k	Range	46 hr, 20 min	56 hr, 57 min	68 hr, 8 min		
Spleen	4.0		.20		17.9	14.2	18.8		
Nervous system								10.74–11.15 (whole brain)	(18)
Cerebrum‡					5.6	5.2	7.2		
Cerebellum‡					7.6	8.2	10.6		
Pons‡					4.8	6.9	8.1		
Medulla‡					6.6	5.8	11.2		
Vagus nerve	.12–.32		14.0–4.2		6.3	5.3	5.9		
Phrenic nerve	.32		4.2	3.90	3.5	3.9			
Spinal cord‡					3.8	4.3	4.7		
Spinal fluid†						1.57			
Dura mater	.32/57*		4.2/.015		3.3	4.2			
Eye									
Whole	.32/24.3*		4.2/.030				2.6		
Cornea	36.0		.022		5.3		10.4		
Sclera and retina	0.32		4.2		4.5	4.7	6.3		
Lens‡					3.7	4.3	5.4		
Aqueous humor‡						1.5	1.9		
Vitreous humor	48.3		.0175		1.5	1.8	2.1		
Optic nerve‡					5.5	8.4	9.7		
Muscle									
Diaphragm	57.0	46.3–68.1	.015	.018–.013		8.1	7.4		
Intercostal muscle	57.0	36.0–68.1	.015	.022–.015	5.4	6.4	6.1		
Tongue	1.5/22.3*	15.7–24.3	.64/.032	.042–.030	11.8	10.6	10.4		
Skeletal muscle	46.3		.018		6.1	3.2	1.9	17.8–36.36	(18–20)
Cartilage	1.5	.32–2.5	.64	4.2–.36	13.0	11.1	22.6		
Tendon or ligament	0.12		14.0		2.9	2.2	3.8		
Tooth‡					8.0	5.7	9.7		
Cortical bone‡					9.9	7.8	13.0	.44% (dry, fat-free)	(18)
Medullary bone	9.3	.32–36.0	.069	4.2–.022					
Bone marrow‡					3.7	15.3	15.7		
Skin	1.5	1.5–4.0	.64	.64–.20	3.8	2.7	4.5	14.87–22.31	(18)
RBC								4.69–5.77§	(21)

* Two distinct peaks found. † Insufficient number of samples. ‡ Continued rise during observation period. § Value in mg/100 ml.

ministered Mg^{28}. From these data the biologic half-life of an intravenously administered dose of Mg^{28} in the dog was calculated to be about 11 days, excluding physical decay.

The time course of calculated total body exchanging Mg^{24} mass and space is summarized in Fig. 6. Because satisfactory measurements of Mg^{24} were available only for plasma, the values were expressed in "plasma equivalents" (Fig. 6). The interrupted line shown in Fig. 6 indicates the average values for extracellular water and total body water per kilogram for the normal dogs (19). It is evident that by 3 hr after intravenous injection the Mg^{28} had distributed itself in a calculated plasma equivalent space which exceeded the total body water. This, of course, reflects the high intracellular concentration of Mg^{24} relative to plasma concentration. The calculated exchanging Mg^{24} mass reached a plateau at a value of approximately 5.2 mEq/kg total body wt.

DISCUSSION

The importance of magnesium in metabolism cannot be overemphasized. Unfortunately, magnesium could not be measured readily and accurately in the body fluids and tissues of plants and animals. The clinical methods have been unreliable, difficult to reproduce, and tedious. The introduction of Mg^{28} radioisotope has

0024177

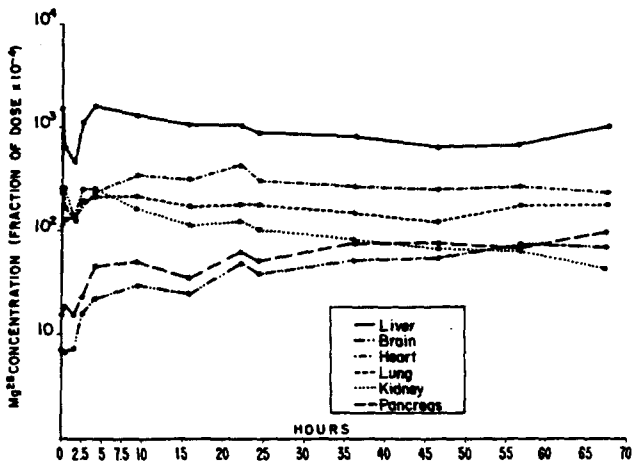

FIG. 5. Concentration-time course of Mg^{28} for various whole organs expressed as a fraction of the dose of Mg^{28} administered.

made possible investigations of the dynamics of magnesium metabolism which were heretofore impossible. Unfortunately the specific activity of the Mg^{28} available for use is still relatively low in spite of recent improvements in its production at Brookhaven National Laboratories. The cost, however, is still extremely high and even prohibitive for most laboratories. In spite of these difficulties these preliminary studies were conducted. Although certain quantitative inaccuracies are known to exist the studies are important from the point of view of the relative role of many tissues in the metabolism of magnesium in an important experimental animal. These studies in the dog are being extended in this laboratory by accurate measurements of magnesium in the frozen and stored tissues of the same dogs using the Zeiss flame spectrophotometer with double monochromater. As methods improve, Mg^{28} of much better specific activity will become available. Further studies will be extended to man. In the meantime, these data for dogs are of interest and certain aspects of the findings are discussed below.

The analysis of the plasma curve into linear exponentials compares fairly well with those obtained by Silver et al. (22) using urinary excretion as the basis for analysis but it does not correspond with the results of others (6, 9, 23). The analysis of composite plasma curves of this type carries a certain degree of arbitrariness not remediable by statistical analysis. The multitude of different apparent rates of exchange and different patterns of uptake in the various sampled tissues serves as some justification for the predication of five (and certainly more) components of the plasma concentration-time course curve. It is likely that the error lies in oversimplification rather than overcomplexity of the analysis of the plasma curve in the complex system of the whole organism.

Figure 4 demonstrates that there are marked differences in the rate of exchange of Mg^{28} by different tissues. Renal tissue showed a rapid rise to a peak during the initial 4 hr and then gradually declined. This was probably due to high radioactivity of the urine excreted by the kidneys initially and the rich supply of blood with high concentration of Mg^{28}. The left ventricle, liver, and pancreas had similar curves of uptake of Mg^{28} except for differences in time at which peak radioactivity was reached. The differences in the rate of accumulation of Mg^{28} among the interventricular septum, left ventricle, and right ventricle noted by others (24) was observed in these dogs through the initial 48 hr. After this time these differences were no longer apparent. Throughout the period of observation the ventricles consistently showed a higher concentration of Mg^{28} than the atria.

TABLE 3. *Comparison of exchanging Mg^{24} in mother and embryos at 68 hr*

Organ	Exchanging Mg^{24} Content, mEq/kg	
	Mother	Embryo[a]
Whole body	4.91[b]	8.4
Brain	7.2[c]	8.4
Heart	23.1[d]	10.9
Lungs	11.9	9.1
Liver	15.7	11.1
Spleen	18.8	12.23
Kidney	5.6[e]	12.45

[a] Average of nine embryos. [b] Based on exchanging mass. [c] Cerebrum. [d] Left ventricle. [e] Average of renal cortex and medulla.

Another interesting finding was the continuing rise in radioactivity in the cerebellum even up to 68 hr (Fig. 4). This was true of all nervous tissue. The concentration of Mg^{28} in skeletal muscle (quadriceps) appeared to reach a peak during the period of observation but there was substantial variability among the animals. It must be remembered that skeletal muscle was at rest, as compared with cardiac muscle which worked continuously. The bladder, predominantly smooth muscle, had a faster rate of exchange than skeletal muscle. Bone and teeth had highly variable isotope concentrations. Both did not reach equilibrium of distribution by 68 hr. The variability seen in radioactive content of muscle and bone apparently represent true physiologic differences and not experimental error.

Although not illustrated, many tissues of high connective tissue content, such as blood vessel walls and pleura, showed two peaks of radioactivity. The first peak, which occurred within the initial 19–90 min, may be interpreted as a reflection of equilibration between plasma with extracellular fluid. Other structures having even larger amounts of connective tissue, such as cartilage, trachea, skin, and tendon, had rapid rates of exchange.

Although the Mg^{28} concentration remained low, the spinal fluid, aqueous humor, and vitreous humor all exceeded the plasma Mg^{28} concentration at the end of the 68 hr of study. This is in part a reflection of the slow rate of exchange of magnesium between that of plasma and these three fluids. Of particular interest also was

0024178

TABLE 4. *Fecal and urinary excretion of Mg^{28} at various time intervals in seven dogs**

Time, hr	Dog No.	Dose, mg Mg^{24}	Urine, % Total Dose Excreted	Feces, % Total Dose Excreted
.32	7	14.8	2.2	
2.5	10	8.3	6.8	
4.05	5	19.8	19.7	
10.9	13	6.9	3.6	1.3
21.8	4	13.96	5.6	.05
22.4	8	14.78	10.3	1.6
23	9	14.78	2.9	2.5
34.7	13	6.9	3.8	2.0
45.8	4	13.96	13.9	3.5
46.3	9	14.78	3.4	4.5
57	13	6.9	8.8	4.1
68.1	4	13.96	20.1	

* The excretion data are cumulative.

the finding that pericardial fluid did not reach plasma levels by the time a final specimen was obtained at 57 hr. Other studies in this laboratory have shown the rate of transfer of Mg^{28} across the pericardium to be slower than the rates for many other ions (T. Takashina, R. Lazzara, J. A. Cronvich, and G. E. Burch, to be published). The presence of relatively high Mg^{28} concentration in bile (as much as 10 times plasma) during the interval from 16 to 68 hr indicates that the liver is an important route of excretion of Mg^{24}. It also reflects the rapid rate of exchange between Mg of plasma and the liver. The extent to which the magnesium excreted in this manner is reabsorbed is not known, but Mg^{28} absorption from the gastrointestinal tract of man has been found to be very slow and small (9, 22). The Mg^{28} concentration in large intestinal contents was higher than small intestinal contents in some animals and lower in others. However, the amount of Mg^{28} lost in the feces was relatively low. The differences must be related not only to the rate of absorption of magnesium but also to the rate of absorption of water.

As would be expected, the Mg^{28} concentration of active glandular tissue (pituitary, adrenals, salivary gland, pancreas) was high, reaching peak levels rapidly. Although Mg^{82} concentration in bone marrow reached levels 11 times that of plasma at 68 hr, bone marrow had not reached a peak level by that time.

The curves of uptake in the various tissues did not all conform with predicted curves based on the model system (Fig. 1). This is not surprising. The possible reasons for this have been discussed and do not require detailed repetition (25). Foremost is the likelihood that one is not sampling single compartments when one samples any tissue. This is a methodological fault and does not necessarily vitiate the model. Second, it is likely that some concatenation of peripheral compartments occurs and some compartments may not exchange directly with the central compartment. For example, it is possible there may be compartments in bone with different rates of exchange as has been demonstrated for calcium. Furthermore, there must be different intracellular pools of Mg^{24} with different exchange rates so that the different intracellular compartments exchange not only with the

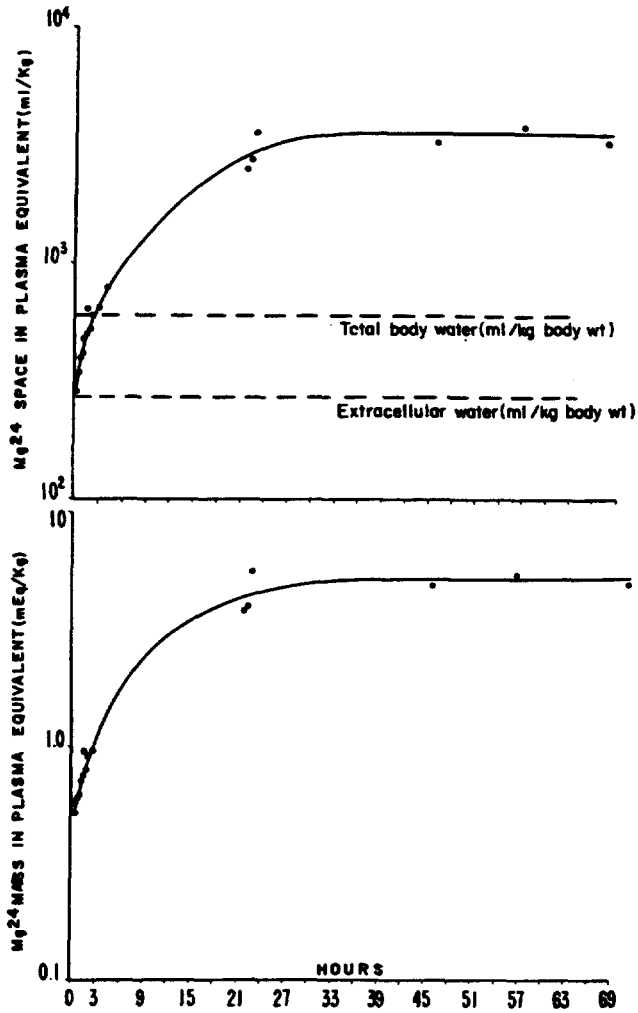

FIG. 6. Mean time course curves of Mg^{24} exchanging space and mass calculated for seven dogs for which satisfactory data were available. The horizontal interrupted lines shown with the curve of exchanging space indicate accepted values (19) for total body water and for extracellular water.

extracellular (or central) compartment, but among themselves. Another special system which can influence the patterns of exchange is the portal system where there are at least two compartments, communicating with the portal compartments which in turn is not uniformly mixed with the whole central compartment being sampled. In view of these complex problems the calculated k values based on the model system outlined for these studies are difficult to interpret and apply. The k values are offered not as numerically exact determinations but as rough impressions reflecting many different dynamic rates of exchange phenomena of the organism.

Because of inadequate data on Mg^{24} content of the tissues of the dog, only a few comparisons could be made with values of calculated exchanging Mg^{24} (Table 2). In some instances (e.g., the heart) the disparity between the calculated values of exchanging Mg^{24} and chemically determined Mg^{24} concentrations reported by others was small. These differences may be due to the 25% over-

0024179

estimation of exchanging Mg^{24} produced by errors due to the lag as well as to errors inherent in the method used by others to measure Mg^{24} in tissue. Muscle (20, 26) and bone (26) have been reported to have concentrations of Mg^{24} measured by chemical methods higher than the concentrations calculated in these experiments to be exchanging Mg^{24} at 68 hr. This is probably due to a large amount of very slowly exchanging magnesium present in these tissues.

The wide variation in calculated concentrations of exchanging Mg^{24} in the organs of the maternal dog contrasts with the uniformity of calculated values of exchanging Mg^{24} in the organs of her embryos (Table 3). The embryos had a higher total exchanging Mg^{24} content per unit body weight than the adult dog, probably because of a lower content of slowly exchanging Mg^{24} pools (i.e., bone Mg^{24}) which contribute relatively little to the exchanging Mg^{24} peak at 68 hr in the adult. Of course this does not imply that the embryos had more chemical Mg^{24} per unit weight, simply that more of the body Mg^{24} had exchanged per unit weight than in the adult at the time the calculations were made.

Reports of the rates of excretion of magnesium in urine after intravenous administration have varied considerably (5, 7, 9, 19, 20) possibly reflecting the influence of the administration of different loading quantities of carrier Mg^{24}. The lower rate of urinary excretion observed in these studies in which Mg^{28} of higher specific activity was used tends to support this possibility (Table 3). Other investigators (5, 22) have noted that the rate of fecal excretion of Mg^{28} is low after intravenous injection, an observation confirmed in these experiments.

The amounts of Mg^{28} excreted in the urine bore no relationship to the dose administered (Table 4) with the possible exception of *dog 5* that received the largest amount of carrier Mg^{24}. This dog had a relatively high rate of urinary excretion. *Dog 7*, receiving the average amount of carrier Mg^{24}, had a plasma Mg^{24} of 1.9 mEq/kg 7 min shortly after injection of the tracer. These considerations indicate that the amounts of carrier Mg^{24} administered were not large enough to produce measurable physiologic effects. Therefore, since tracer conditions were at least approximated, the estimate of biologic half-life in these experiments was probably reasonably accurate.

The finding that the plasma equivalent Mg^{24} space exceeded the total body water in less than 3 hr (Fig. 6) is consistent with the fact that Mg^{24} was highly concentrated in the intracellular compartment and equilibrium of distribution had not been achieved. Other investigators (19) have reported similar observations. The occurrence of a plateau in this curve at 24–28 hr does not indicate that no further cellular uptake was in progress but that the uptake was either too small or too slow to be detected by the methods employed. There was direct evidence that tracer was still increasing in concentration in some organs (e.g., brain) at 68 hr. Our findings confirm those of others (9) in that the calculated total exchanging Mg^{24} mass at the end of the period of observation (5.2 mEq/kg) was substantially less than that found by chemical analysis of tissues of mammalian species (1, 2). The explanation for this difference is evident from the great differences between calculated exchanging Mg^{24} and chemically determined concentration of Mg^{24} in muscle and bone (vide supra). Because of their large bulk these tissues probably account for the largest part of the Mg^{24} which still was not exchanging within 68 hr of the time the tracer was administered.

REFERENCES

1. Barker, E. S. *J. Chronic Diseases* 11: 278, 1960.
2. Wacker, W. E. C., and B. L. Valee. *New Engl. J. Med.* 259: 431, 475, 1958.
3. Elkington, J. R. *Clin. Chem.* 3: 319, 1957.
4. MacIntyre, I. *Proc. Roy. Soc. Med.* 52: 212, 1959.
5. Brandt, I. L., W. Glaser, and A. Jones. *Metabolism* 7: 355, 1958.
6. Rogers, T. A., and P. E. Mahan. *Proc. Soc. Exptl. Biol. Med.* 100: 235, 1959.
7. Glicksman, A. S., M. K. Schwartz, H. Bone, K. E. Roberts, and H. T. Randall. *Clin. Res.* 4: 14, 1956.
8. Aikawa, J. K., E. L. Rhodes, A. L. Hanns, and J. L. Reardon. *Am. J. Physiol.* 197: 99, 1959.
9. Aikawa, J. K., G. S. Gordon, and E. Rhodes. *J. Appl. Physiol.* 15: 325, 1960.
10. Aikawa, J. K. *Proc. Soc. Exptl. Biol. Med.* 103: 363, 1960.
11. Aikawa, J. K. *Proc. Soc. Exptl. Biol. Med.* 104: 461, 1960.
12. Love, W. D., R. B. Romney, and G. E. Burch. *Circulation Res.* 2: 112, 1954.
13. Walsh, J. J., and G. E. Burch. *J. Lab. Clin. Med.* 54: 59, 1959.
14. Sheppard, C. W., and W. R. Martin. *J. Gen. Physiol.* 33: 703, 1950.
15. Alcock, N., I. MacIntyre, and I. Radde. *J. Clin. Pathol.* 13: 506, 1960.
16. Threefoot, S. A., C. T. Ray, and G. E. Burch. *J. Lab. Clin. Med.* 45: 395, 1955.
17. Threefoot, S. A., G. E. Burch, and P. Reaser. *J. Lab. Clin. Med.* 34: 1, 1949.
18. Spector, W. S. (editor). *Handbook of Biological Data.* Philadelphia: Saunders, 1956, pp. 70–77.
19. Harrison, H. E., D. C. Darrow, and H. Yannet. *J. Biol. Chem.* 113: 515, 1936.
20. Eichelberger, L., and F. C. McLean. *J. Biol. Chem.* 14[illegible] 467, 1942.
21. Somes, D. A., W. F. McGuckin, and K. G. Wakins. *Clin. Chem.* 6: 564, 1960.
22. Silver, L., J. S. Robertson, and L. K. Dahl. *J. Clin. Invest.* 39: 420, 1960.
23. Zumoff, B., E. H. Bernstein, J. J. Imarisio, and L. Hellman. *Clin. Res.* 6: 260, 1958.
24. Glover, W., and J. L. Brandt. *Am. J. Physiol.* 196: 375, 1959.
25. Ginsberg, J. M., and W. S. Wilde. *Am. J. Physiol.* 179: [illegible] 1954.
26. MacIntyre, I. *Proc. Roy. Soc. Med.* 53: 1037, 1960.

0024180