THE RENAL EXCRETION OF INORGANIC PHOSPHATE IN RELATION TO THE ACTION OF VITAMIN D AND PARATHYROID HORMONE[1]

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The mode of action of vitamin D in the prevention and cure of rickets is still not understood. In 1921, Howland and Kramer (1) pointed out that the concentration of phosphorus in the serum was usually reduced in human rickets, at times to very low levels, although the concentration of calcium might be within normal limits. This has been confirmed repeatedly and it has been found that the administration of vitamin D results in a prompt rise in the concentration of phosphorus, even to abnormally high levels with excessive dosage. The mechanism through which vitamin D influences the concentration of phosphorus in the body fluids has not been satisfactorily explained. In balance experiments in man (2, 3) and experimental animals (4) it has been shown that during states of vitamin D deficiency absorption of calcium and phosphate from the intestinal tract is reduced and that the administration of vitamin D increases the absorption of these ions. In the rat, the studies of Nicolaysen (5) have indicated that in the absence of vitamin D the absorption of calcium is primarily diminished and that the absorption of phosphate is only secondarily affected. Careful examination of the data from metabolic studies in the rachitic infant leads to the conclusion that deficient absorption of phosphate from the intestinal tract cannot wholly explain the diminished concentration of phosphate in the plasma. In infants developing rickets on a cow's milk diet, *i.e.*, a diet high in both calcium and phosphorus, the amount of phosphate absorbed from the intestinal tract would be sufficient for the needs of the infant were the phosphate retained rather than excreted in the urine.

The excretion of phosphate in the urine must play an important rôle in the regulation of the concentration of phosphate in the body fluids. How is the renal excretion of phosphate controlled and does vitamin D influence the concentration of phosphate in the blood plasma through an effect on the excretion of this ion by the kidneys? The present experiments were conducted to answer these questions.

The Renal Excretion of Phosphate

Walker (6) and Walker and Hudson (7) have shown that the phosphate of the plasma is filtrable through the glomerular membranes of the amphibian. These workers have also found that under certain conditions active reabsorption of phosphate takes place in the proximal tubules so that the urine in the distal tubules and bladder may be almost phosphate-free. There was no evidence that phosphate was secreted into the lumen of the tubules.

In the dog, Pitts (8) has shown that the clearance of phosphate increases as the concentration of phosphate in the plasma increases and approaches the xylose clearance at very high levels of plasma phosphate. At normal levels of phosphate in the blood plasma the urine may be almost phosphate-free and the excretion of phosphate in the urine is only a small fraction of that filtered through the glomeruli. In the mammalian kidney, therefore, reabsorption of phosphate must also take place as the glomerular filtrate passes through the renal tubules.

In order to study this tubular reabsorption of phosphate quantitatively, simultaneous determinations of the creatinine and phosphate clearances were made in the dog following the intravenous injection of phosphate salts.

METHODS

Female dogs fed standardized diets were used for the experiments. The studies of phosphate excretion were made about 18 to 20 hours after the last feeding. The

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animals were unanesthetized and lay comfortably on the animal board with loose restraints. A solution of 0.1 molar sodium phosphate of approximately pH 7.4 was injected intravenously in a dosage of 7 to 10 cc. per kilogram of body weight, together with creatinine (0.3 gram per kgm.). In most of the experiments 300 cc. of water were given by stomach tube in order to increase the urine volume, thus making the collections more accurate. The animals were catheterized and urine was collected quantitatively during successive periods of 10 to 20 minutes each. The bladder was washed with measured volumes of distilled water at the end of each period. Repeated blood samples were taken from the external jugular vein during the periods of urine collection. All analyses were done on the separated serum and the same analytical methods were used for serum and urine. Creatinine was determined by the usual Jaffé reaction (9); inorganic phosphate, by the Fiske and Subbarow method (10). A photoelectric colorimeter was used.

In order to calculate the clearances of creatinine and phosphate, the concentrations as determined in the serum were plotted on semi-logarithmic paper and the concentrations at the mid-point of each period obtained from the curve. The plasma clearances were then calculated by the usual formula $C = \frac{UV}{P}$. It has been demonstrated that the creatinine clearance in the dog can be used as a measure of the rate of glomerular filtration (11). If the plasma phosphate is completely filtrable through the glomeruli, the differences between the clearances of creatinine and those of phosphate measure the rate of reabsorption of phosphate by the renal tubules.

Following the injection of phosphate salts it has been shown, however, that a colloidal phosphate complex is formed which is not filtrable through collodion membranes (12). This non-ultrafiltrable fraction of the phosphate rapidly disappears from the plasma if no further phosphate is given. Repeated studies of the filtrability of the serum phosphate through collodion membranes were made following the intravenous injection of sodium phosphate in the amounts described above. The membranes were prepared by the method of Greenberg and Gunther (13) and the apparatus used for ultrafiltration was that described by Benjamin and Hess (14). The results of such experiments are shown in Table I.

TABLE I

Filtrability of plasma inorganic phosphate through collodion membranes following intravenous injection of sodium phosphate

Experiment	Serum calcium	Time elapsed	Phosphate		Filtrable
			Plasma water	Ultrafiltrate	
	mgm. per 100 cc.	*minutes*	*mgm. per 100 cc.*		*per cent*
1	9.0	82	10.8	9.9	92
		163	9.9	9.8	99
2	10.5	21	9.2	8.1	88
		50	7.0	6.9	99
		95	6.1	5.9	97
		144	6.2	5.8	94
3*	11.9	63	8.4	8.0	95
		97	5.9	6.0	100
4*	14.6	63	8.6	7.2	84
		97	6.5	6.1	94
		136	5.9	6.0	100
		177	5.3	5.3	100
5†	14.5	31	10.6	8.0	76
		71	6.9	6.7	97
		111	6.5	6.1	95
6‡	16.0	63	7.4	6.5	88
		96	5.6	4.9	89
		134	5.0	5.1	100
		178	4.9	5.0	100

* Following administration of vitamin D.
† Following administration of dihydrotachysterol (A. T. 10).
‡ Parathyroid extract 2 cc. injected preceding day.

It may be seen that soon after the concentration of phosphate in the plasma is raised by the intravenous injection, an appreciable fraction of the inorganic phosphate is not filtrable through collodion. In about 60 to 90 minutes, however, the phosphate is again completely or almost completely filtrable. This is true even in the experiments in which the serum calcium was raised above the normal value by the administration of vitamin D or parathyroid extract. In the studies of renal function, the determinations of the clearances of phosphate were started 60 minutes following the intravenous injection of phosphate salts and continued for about 2 hours. After this interval the plasma phosphate is essentially completely filtrable through collodion membranes and it is assumed that it is also completely filtrable through the glomerular membranes.

The results of typical experiments are shown in Tables II, III and IV. The creatinine clearances are seen to remain relatively constant in the successive periods. The clearances of phosphate, however, decrease as the concentration of phosphate in the plasma decreases and in some cases the excretion of phosphate in the urine may be reduced almost to zero. If the creatinine clearance be taken as the rate of glomerular filtration, since the concentration of phosphate in the plasma is known, the amount of phosphate filtered through the glomeruli per minute may be calculated. The quantity of phosphate reabsorbed by the renal tubules is obtained by subtraction of the

TABLE II

The reabsorption of phosphate by the renal tubules

(Dog B. Weight 9.6 kgm.)

Period	Total elapsed time	Urine volume	Creatinine clearance	Plasma phosphate	Phosphate clearance	Phosphate		
						Filtered	Excreted	Reabsorbed
	minutes	*cc. per minute*	*cc. per minute*	*mgm. per 100 cc.*	*cc. per minute*	*mgm. per minute*		
	0	400 cc. water by stomach tube						
1	60–70	1.9	35.0	4.6	5.7	1.61	0.26	1.35
	72–80	125 cc. 0.1 M phosphate intravenously						
2	120–140	1.8	38.5	9.4	23.2	3.62	2.18	1.44
3	140–160	0.55	31.0	7.3	15.5	2.26	1.13	1.13
4	160–170	0.8	36.0	6.6	17.0	2.38	1.12	1.26

TABLE III

The reabsorption of phosphate by the renal tubules

(Dog A. Weight 7.7 kgm.)

Period	Total elapsed time	Urine volume	Creatinine clearance	Plasma phosphate	Phosphate clearance	Phosphate		
						Filtered	Excreted	Reabsorbed
	minutes	*cc. per minute*	*cc. per minute*	*mgm. per 100 cc.*	*cc. per minute*	*mgm. per minute*		
1	0–10		23.5	3.3	2.4	0.78	0.08	0.70
	12–20	100 cc. 0.1 M phosphate intravenously						
2	70–90	1.45	26.2	6.5	14.0	1.70	0.91	0.79
3	90–110	0.45	22.3	5.5	9.8	1.23	0.54	0.69
4	110–130	0.25	22.7	5.0	8.8	1.14	0.44	0.70
5	130–150	0.20	22.6	4.6	7.4	1.04	0.34	0.70
6	150–170	0.20	23.9	4.5	6.0	1.08	0.27	0.81
7	170–190	0.20	23.0	4.4	5.0	1.01	0.22	0.79

amount excreted in the urine from the amount filtered. If such calculations are made, it may be seen that the tubular reabsorption of phosphate expressed as milligrams of phosphorus per minute is essentially constant and is not influenced by the elevation of the concentration of phosphate in the serum. In many of the experiments the calculated tubular reabsorption of phosphate is found to vary to some extent with fluctuations in the creatinine clearance, suggesting that the reabsorption of phosphate is in part affected by changes in the filtration rate. The present experiments cannot answer this point since the tubular reabsorption of phosphate is determined indirectly. Any errors in the determination of the creatinine clearance would produce an error in the same direction in the calculation of the phosphate reabsorption.

If the tubular reabsorption of phosphate remains constant as the concentration of phosphate in the plasma decreases, a concentration should be reached at which the quantity filtered equals the quantity reabsorbed and no phosphate should be excreted in the urine. This estimated concentration of phosphate will be termed the equilibrium concentration and may be calculated by the following formula: $C_E = \frac{T_m}{F} \times 100$, where C_E is the equilibrium concentration, T_m is the maximal rate of reabsorption of phosphate and F, the rate of glomerular filtration. The reabsorption of phosphate by the renal tubules may not be complete and traces may be present in the urine at concentrations of phosphate in the plasma below the calculated equilibrium concentration. In the dog, at least, in those experiments in which the concentration of phosphate in the plasma decreased to the calculated equilibrium value, the excretion of phosphate in the urine dropped to less than 0.002 milligrams per minute, indicating that less than 0.1 per cent of the phosphate filtered escaped reabsorption.

The rate of reabsorption of phosphate is not affected by water diuresis. In these experiments

TABLE IV

The reabsorption of phosphate by the renal tubules

(Dog C. Weight 9.9 kgm.)

Period	Total elapsed time	Urine volume	Creatinine clearance	Plasma phosphate	Phosphate clearance	Phosphate		
						Filtered	Excreted	Reabsorbed
	minutes	*cc. per minute*	*cc. per minute*	*mgm. per 100 cc.*	*cc. per minute*	*mgm. per minute*		
	EXPERIMENT I							
	0–10	75 cc. 0.1 M phosphate intravenously						
1	50–70	0.45	43.3	9.8	7.6	4.24	0.74	3.50
2	70–90	0.9	42.3	9.4	5.1	3.97	0.48	3.49
3	90–110	1.9	43.4	9.2	5.0	3.99	0.46	3.53
4	110–130	1.8	43.1	9.0	4.1	3.88	0.37	3.51
5	130–150	1.3	40.6	8.9	2.6	3.61	0.23	3.38
	EXPERIMENT II							
	0–10	75 cc. 0.1 M phosphate intravenously						
1	55–75	0.1	39.5	9.5	3.1	3.75	0.29	3.46
2	95–115	0.2	38.7	9.0	1.2	3.48	0.11	3.37
3	115–135	0.15	35.7	8.9	0.5	3.18	0.04	3.14
4	135–155	0.35	41.3	8.8	0	3.63	0	3.63

the rate of urine excretion was varied from 0.1 to 5 cc. per minute without significant changes in the tubular reabsorption of phosphate. This is in accord with previous reports that the urinary excretion of phosphate is not increased by water diuresis (15).

In experiments in the young dog extending over many months, variations in the renal clearances of phosphate are observed which are apparently related to age. The concentration of plasma inorganic phosphate in the young dog is much higher than that in the adult animal. This is similar to the findings reported in man. Anderson and Elvehjem (16) have also recently found that the concentration of plasma inorganic phosphate in the dog decreases with age. Comparative studies of the creatinine clearances and renal tubular reabsorption of phosphate in dogs from 2 to 27 months of age are shown in Table V. In the young animal the rate of tubular reabsorption of phosphate is greater in proportion to the rate of glomerular filtration than in the adult dog. In dog C during the latter part of the first year of life, at which time the filtration rate has reached a relatively constant level, there is a decrease in the rate of tubular reabsorption of phosphate from the maximum values, and the calculated equilibrium concentration of phosphate decreases markedly. The data for dog B indicate that there is a gradual decrease in the rate of tubular reabsorption of phosphate during the second year of life, with no change in the filtration rate. In most of the experiments the concentrations of phosphate in the fasting state approximate the calculated equilibrium concentration.

TABLE V

The influence of age upon the concentration of phosphate in the serum and the renal tubular reabsorption of phosphate

Dog	Age	Weight	Creatinine clearance	Phosphate reabsorbed	Equilibrium concentration	Serum * phosphorus
	months	*kgm.*	*cc. per minute*	*mgm. per minute*	*mgm. per 100 cc.*	
A	4	5.7				9.6
	17	7.7	23.3	0.94	4.0	4.7
B	4					7.3
	13	9.0	31.1	1.43	4.6	4.9
	20	9.7	29.9	1.25	4.2	4.6
	27	9.7	32.9	1.04	3.2	3.4
C	3	7.7	29.1	2.30	7.9	7.7
	7	9.9	42.5	3.48	8.2	8.2
	8	10.6	43.9	3.46	7.9	7.7
	10	10.9	43.2	2.66	6.2	6.7
	12	10.7	38.2	1.79	4.7	5.9
D	2½	8.5	44.0	4.22	9.6	8.4

* Blood taken 18 hours after last feeding.

The effect of vitamin D upon the renal excretion of phosphate

A 6-week old female collie puppy was placed on a vitamin D free diet, low in both calcium and phosphorus. The composition of the diet was a modification of that described by Morgan (17). At the age of 20 weeks rachitic changes in the bones could be demonstrated by roentgenogram. The concentrations of calcium and phosphorus in the serum were both reduced. During the period of active rickets repeated studies of the tubular reabsorption of phosphate were carried out as described in the preceding section. The animal was then given 20,000 units vitamin D in the form of irradiated ergosterol each day for 3 days, totaling 60,000 units. On the fourth day, studies of the renal function were done and repeated at intervals of 1 to 2 weeks thereafter

TABLE VI

The effect of vitamin D upon the renal tubular reabsorption of phosphate in the rachitic dog

Date	Creatinine clearance	Phosphate reabsorbed	Equilibrium concentration	Serum	
				Phosphorus	Calcium
	cc. per minute	*mgm. per minute*	*mgm. per 100 cc.*	*mgm. per 100 cc.*	
1939					
October 2.....	Rachitogenic diet started—age 6 weeks				
October 26....				8.8	10.6
November 29 .	29.1	2.30	7.9	7.7	12.7
December 22..	37.9	2.56	6.8	6.2	8.5
1940					
January 8....	43.1	2.40	5.6	5.7	8.4
January 22...	45.4	2.56	5.6	6.0	7.6
February 12..	20,000 units vitamin D				
February 13..	20,000 units vitamin D				
February 14..	20,000 units vitamin D				
February 15..	40.7	3.87	9.5	9.0	9.0
February 28..	43.1	3.47	8.1	8.2	10.5
March 27.....	44.1	3.46	7.9	7.4	9.6
April 25.....	45.0	3.19	7.1	7.2	8.5
June 3.....	50.2	3.12	6.2	6.6	8.6
June 16.....	80,000 units vitamin D				
June 17.....	49.9	3.49	7.0	6.6	9.3
June 28.....	45.3	2.91	6.4	6.7	10.2

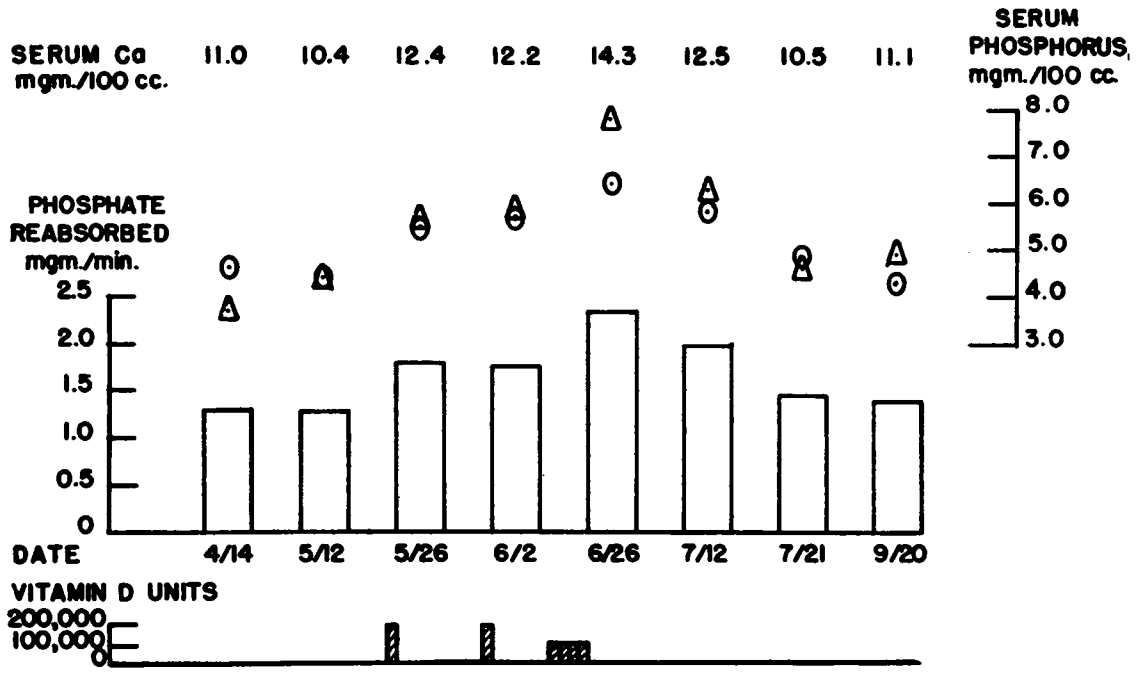

FIG. 1. THE EFFECT OF VITAMIN D ON TUBULAR REABSORPTION OF PHOSPHATE—MATURE DOG B

Columns represent the average tubular reabsorption of phosphate—expressed as mgm. of phosphorus per minute.

⊙ Concentration of serum phosphorus in fasting state.

△ Calculated "equilibrium concentration" of phosphate expressed as mgm. phosphorus per 100 cc.

without any further treatment with vitamin D. The results are shown in Table VI. In this and succeeding tables, the data for each experiment represent the average of 5 or 6 successive periods of 20 minutes each.

It is evident that following the administration of vitamin D, there is a marked increase in the rate of reabsorption of phosphate by the renal tubules. The creatinine clearances remain unchanged. The calculated equilibrium concentration of phosphate is therefore increased from 6 mgm. per 100 cc. during the rachitic state to 9.5 mgm. per 100 cc. following treatment with vitamin D. The concentrations of phosphorus in the serum during the postabsorptive state parallel closely this calculated value. The data indicate that in these experiments the effect of vitamin D in raising the concentration of phosphorus in the serum is due to its action on the tubular reabsorption of phosphate, preventing loss of phosphate in the urine. The effect of the treatment with 60,000 units of vitamin D is seen to persist for several months with a gradual decrease in the rate of tubular reabsorption of phosphate, until the pretreatment level was reached. At this time further treatment with vitamin D was again effective in increasing the reabsorption of phosphate by the renal tubules, but to a much slighter extent than previously.

A second dog was studied in similar manner except that the animal was not started on the vitamin D free diet until 3 or 4 months of age and no evidences of rickets were demonstrated by roentgenogram. The animal had already become sexually mature before the renal function studies were done. Following the administration of vitamin D, however, an increase in the reabsorption of phosphate by the renal tubules was also found with an associated increase in the concentration of phosphate in the serum as shown in Figure 1. This effect could be demonstrated 24 hours after the animal had been given a single large dose of vitamin D (200,000 units). Following the administration of a total of 800,000 units of vitamin D, a still greater effect upon the renal tubular reabsorption of phosphate was found. With this

dosage a marked hypercalcemia developed. Following cessation of treatment, the renal tubular reabsorption of phosphate rapidly fell to the previous levels and the concentration of serum phosphorus in the fasting state likewise decreased. With the dosage of vitamin D used no effect upon the creatinine clearances was noted. However, in another dog given 1,600,000 units of vitamin D over a period of 8 weeks, the creatinine clearance decreased from approximately 25 cc. per minute to 15 cc. per minute. This effect upon glomerular filtration was not apparently associated with permanent injury to the kidney since the creatinine clearance again returned to normal levels following cessation of treatment. With the administration of excessive amounts of vitamin D to dogs, Goormaghtigh and Handovsky (18) have described anatomical changes in the kidney and hypertension. Although vitamin D is extremely effective in increasing the tubular reabsorption of phosphate in the growing dog depleted of vitamin D, only a slight effect may be produced upon the renal tubular reabsorption of phosphate in the normal adult animal, even though extremely large amounts, sufficient to raise the serum calcium to abnormally high levels, are given. In agreement with these observations are the reports that in the adult human as well as in the adult dog the concentration of phosphorus in the plasma is increased but slightly following the administration of vitamin D (19).

The effect of parathyroid extract on renal tubular reabsorption of phosphate

Many workers have attempted to correlate the physiological actions of vitamin D with those of the parathyroid hormone. Although both vitamin D and parathyroid extract produce an increase in the concentration of calcium in the serum, their effects upon the concentration of phosphate are diametrically opposite. Studies of the effects of parathyroid extract upon renal tubular reabsorption of phosphate are shown in Tables VII and VIII.

In each of the experimental animals a marked decrease in the renal tubular reabsorption of phosphate was found following the injection of parathyroid extract. No change in the creatinine clearance was noted with the amount of parathyroid extract used in these experiments. The calculated equilibrium concentrations of phosphate were markedly decreased from the control values. This effect of parathyroid extract could also be demonstrated in the rachitic dog, although relatively large amounts of extract were necessary to affect the renal excretion of phosphate (Table VIII). It may be seen that the already low rate of renal tubular reabsorption of phosphate in the rachitic dog is decreased still further with an associated decrease in the concentration of serum phosphorus. In contrast to the effect of para-

TABLE VII

Effect of parathyroid extract on renal tubular reabsorption of phosphate

Treatment	Creatinine clearance	Phosphate reabsorbed	Equilibrium concentration	Serum	
				Phosphorus	Calcium
	cc. per minute	*mgm. per minute*	*mgm. per 100 cc.*	*mgm. per 100 cc.*	
Dog A					
Control.....	23.5	0.89	3.8	4.1	11.0
Parathyroid extract 2 cc.*	25.2	0.57	2.3	3.0	14.1
Dog B					
Control.....	30.9	1.37	4.4	4.6	10.5
Parathyroid extract 4 cc.*	30.9	0.89	2.9	4.0	14.7

* Given subcutaneously in divided doses the day preceding the experiment.

TABLE VIII

The effect of parathyroid extract on the renal tubular reabsorption of phosphate in rachitic dog

Date	Treatment	Creatinine clearance	Phosphate reabsorbed	Equilibrium concentration	Serum	
					Phosphorus	Calcium
		cc. per minute	*mgm. per minute*	*mgm. per 100 cc.*	*mgm. per 100 cc.*	
January 22	Control	45.4	2.56	5.6	6.0	7.6
January 30	Parathyroid extract 6 cc.*	43.3	2.00	4.6	5.3	9.1
February 8	Control	42.5	2.55	6.0	6.2	8.4
February 15	Vitamin D 60,000 units	40.7	3.87	9.5	9.0	9.0

* Given in divided doses during day preceding experiment.

thyroid extract is shown the result of treatment with vitamin D, as previously described. The observed decrease in the rate of reabsorption of phosphate by the renal tubules following the injection of parathyroid extract explains the repeated observation of the increased urinary excretion of phosphate and diminished concentration of phosphate in the serum produced by treatment with parathyroid extract and in states of hyperparathyroidism. Conversely, the very high concentrations of phosphorus in the serum in hypoparathyroidism are presumably the result of increased tubular reabsorption of phosphate. When excessive amounts of parathyroid extract are injected, a marked decrease in the creatinine clearance may be observed with a return to normal following cessation of treatment. This effect of toxic doses of parathyroid extract in diminishing glomerular filtration may explain the secondary rise in serum phosphorus when excessive doses of parathyroid extract are injected. With a considerable decrease in glomerular filtration and the liberation of large amounts of phosphate from bone, the serum phosphorus may rise even though tubular reabsorption of phosphate is reduced.

DISCUSSION

"The kidneys appear to serve as the ultimate guardians of the constitution of the internal environment, which they maintain with increasing accuracy under most unfavorable circumstances." Peters (20) has thus emphasized the activities of the kidneys in preventing loss of essential solutes from the body fluids as well as in the elimination of substances present in excess. These observations apply to the function of the kidney in maintaining the concentrations of inorganic phosphate in the plasma and other body fluids. The quantity of phosphate filtered through the glomeruli per day is greatly in excess of the phosphate intake. However, the reabsorption of most of the phosphate from the glomerular filtrate as it passes through the renal tubules prevents the loss of excessive quantities of phosphate in the urine and thus allows for the maintenance of the normal concentrations of phosphate in the body fluids.

The experiments reported here demonstrate that under given conditions there is a maximal rate of tubular reabsorption of phosphate. When the concentration of phosphate in the plasma is increased by the administration of phosphate salts, the quantity of phosphate filtered in excess of the reabsorptive capacity of the tubules is excreted in the urine. Shannon and Fisher (21) first demonstrated this phenomenon of a limiting maximal rate of tubular reabsorption in connection with the reabsorption of glucose and other sugars. Shannon (22) has postulated the theory that an intermediate compound is formed in the renal tubule cells which again dissociates, liberating the free solute into the body fluids, and that the rate of this reaction is the limiting factor in the reabsorption of certain solutes by the tubule cells.

The importance of the renal excretion of phosphate in regulating the concentration of this ion in the body fluids is shown clearly by the experiments reported here. The concentration of phosphate may, of course, be influenced by many factors other than the renal excretion, *e.g.*, the availability of phosphate for absorption from the gastro-intestinal tract, the movement of phosphate from the extracellular fluids into the cells or vice versa, the precipitation of calcium phosphate in the skeleton, or the mobilization of phosphate from the bones into the body fluids. At equilibrium, however, the concentration of phosphate in the plasma approaches the concentration at which the rate of reabsorption of phosphate by the renal tubules is approximately equal to the rate at which phosphate is filtered through the glomeruli.

It has been shown that following the administration of vitamin D there is a rapid increase in the tubular reabsorption of phosphate, the rate of glomerular filtration remaining unchanged. The increased concentration of phosphate in the body fluids produced by the administration of vitamin D can be explained as the result of this effect upon renal tubular function. The work of other investigators has shown that vitamin D influences the absorption of calcium, and secondarily of phosphate, from the intestinal tract. This action of vitamin D in conjunction with its effects upon renal function would result in sustained high concentrations of both calcium and phosphate in the body fluids, a condition favorable for rapid calcification. It is probable that the antirachitic potency of vitamin D is dependent on this com-

bined action on renal tubular function and intestinal absorption.

In this connection, cases of rickets which do not respond to vitamin D therapy are of considerable interest. One group of such cases has been reported (23) in which the concentration of phosphate in the serum is exceedingly low and is not increased by the administration of vitamin D. Studies of these patients reveal marked loss of phosphate in the urine, despite low concentrations of this ion in the blood plasma. These patients may also exhibit other evidences of renal dysfunction, such as renal glycosuria. Albright, *et al* (24) have reported a case of low phosphorus rickets refractory to treatment with vitamin D and associated with diffuse calcification of the kidneys. It is possible that the development of low phosphorus rickets in these cases is due to a failure of the renal tubular mechanisms concerned with the reabsorption of phosphate and that this tubular deficiency is not corrected by the administration of vitamin D. It is of further interest that many of these patients exhibit a severe chronic acidosis. Preliminary studies in the dog have shown that following the production of acidosis, the renal tubular reabsorption of phosphate is greatly diminished (25).

The demonstrated effect of parathyroid extract in diminishing the reabsorption of phosphate by the renal tubules is in agreement with earlier observations that parathyroid extract produces an increase in the excretion of phosphate in the urine, with a simultaneous decrease in the concentration of phosphate in the plasma. This effect may also be related to the changes found in rickets. Hyperplasia of the parathyroids has been observed postmortem in cases of severe rickets (26), and Hamilton and Schwartz (27) have found evidence of an increased amount of parathyroid hormone in the blood of rachitic rabbits. Albright and Sulkowitch (28) have suggested that the low serum phosphorus found in rickets may be due to the secondary hyperparathyroidism present. The hyperplasia of the parathyroids in rickets has been explained as a compensatory response to the calcium deficiency which results from the failure of the intestinal tract to absorb the calcium. Recently Ham *et al* (29) have found that enlargement of the parathyroids could be produced in rats by feeding diets low in calcium and deficient in vitamin D. However, when given a high calcium vitamin D free diet, the rats did not show enlargement of the parathyroids although they did develop low phosphorus rickets. In the experiments reported here the prompt increase in the tubular reabsorption of phosphate observed following the administration of vitamin D suggests that vitamin D exerts a direct effect upon the renal tubule cells.

SUMMARY

By means of the concurrent determinations of creatinine and phosphate clearances in the dog, following the intravenous injection of phosphate salts, it is possible to study quantitatively the reabsorption of phosphate by the renal tubules. It is found that under standard conditions there is a limiting maximal rate of reabsorption of phosphate by the renal tubules which does not vary when the concentration of phosphate in the plasma is elevated by the administration of phosphate salts. The phosphate filtered through the glomeruli which is in excess of the maximum which can be reabsorbed by the renal tubules is excreted in the urine.

The administration of vitamin D to young dogs who have been fed a rachitogenic diet produces a marked increase in the maximal rate of reabsorption of phosphate by the renal tubules, thus increasing the concentration of inorganic phosphate in the plasma at equilibrium. This effect is demonstrable 24 hours after adequate amounts of vitamin D are given and is probably an important factor in its antirachitic activity.

The effect of parathyroid extract upon the tubular reabsorption of phosphate is opposite to that of vitamin D. Following injections of parathyroid extract there is a considerable decrease in the rate of reabsorption of phosphate by the renal tubules and a consequent reduction in the concentration of phosphate in the plasma.

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