creased deposition in the bones is not wholly unexpected. Such phenomena are observed in the deposition of inorganic bone normally. As a rule, the higher the concentration of the bone forming elements in the blood plasma the greater the ease of deposition. Lead in this case may be considered as one of the bone-forming elements, *i. e.*, a part of the inorganic matter of bone, although not a normal one. The mechanism of deposition of lead salts seems to follow the phenomena observed with the normal bone-forming constituents, namely, calcium and phosphate, *i. e.*, the higher the value of the product formed by multiplying the concentrations of these two constituents in the blood serum, the greater the ease of deposition. Another parallel worth mentioning is that vitamin D, as a rule, is instrumental in raising the above product and thereby promotes the normal formation of inorganic bone, just as it promotes deposition of lead in the bone.

Conclusions. Vitamin D causes a rise in the concentration of lead in the blood stream and in the bones of rats suffering from lead poisoning.

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Influence of Dietary Calcium and Phosphorus upon Action of Vitamin D in Experimental Lead Poisoning.

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In the preceding paper[1] a marked effect of vitamin D on the blood and bone lead concentrations of lead-fed animals was demonstrated. In view of the great importance attached to the dietary calcium and phosphorus content in lead poisoning[2, 3, 4] investigations were un-

1 Sobel, A. E., Gawron, O., and Kramer, B. K., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 433.

2 Shelling, D. H., PROC. SOC. EXP. BIOL. AND MED., 1932, **30**, 248; Shelling, D. H., *The Parathyroids in Health and in Disease*, St. Louis, 1935, C. V. Mosby Company; Shelling, D. H., and Hopper, K. B., *Bull. Johns Hopkins, Hosp.*, 1936, **58**, 137.

3 Gray, I., *J. Am. Med. Assn.*, 1935, **104**, 200.

4 Aub, J. C., *J. Am. Med. Assn.*, 1935, **104**, 87; Aub, J. C., Fairhall, L. T., Minot, A. S., and Reznikoff, P., *Lead Poisoning-Medicine Monographs*, Baltimore, Williams and Wilkins Co., 1926, 7.

dertaken to determine the part these factors play upon the previously noted action of the antirachitic vitamin.

The basal diet used in the previous experiment contained traces of calcium (0.03%) and small amounts of phosphorus (0.246%) to which basic lead carbonate was added. To this basal diet 2.5% of calcium carbonate was added to produce a high calcium low phosphorus diet; and 2.75% of anhydrous Na_2HPO_4 was added to produce the high phosphorus low calcium diet. The diets are described below:

		Parts
Basal Diet:	Yellow corn meal (Quaker Oats)	70
	Wheat gluten	16
	Brewer's Yeast (Mead Johnson)	10
	NaCl	1
Diet A:	Basal diet + 3 parts of $Pb(OH)_2 \cdot 2PbCO_3$	
	Pb = 2.4 % = 11.6 mg. mol./100 g.	
	Ca = 0.03 % = .8 " " "	
	P = 0.246% = 7.9 " " "	
Diet B:	Basal diet + 0.5 part of $Pb(OH)_2 \cdot 2PbCO_3$ + 2.5 parts of $CaCO_3$	
	Pb = 0.4 % = 1.93 mg. mol./100 g.	
	Ca = 1.0 % = 25.0 " " "	
	P = 0.246% = 7.9 " " "	
Diet C:	Basal diet + 1.5 parts of $Pb(OH)_2 \cdot 2PbCO_3$ + 2.75 Na_2HPO_4	
	Pb = 1.2 % = 5.8 mg. mol./100 g.	
	Ca = 0.03 % = .8 " " "	
	P = 0.846% = 27.2 " " "	

Albino rats raised in our laboratory from an original Wistar strain were used. The mothers were kept on the stock diet of Bills, *et al.*[5] The young were weaned at 21 days, at which time they were placed on the stock diet. At the age of 23 to 25 days this was replaced by the experimental diets described above. One half of each group was given 33 Steenbock units of vitamin D. (Mead Johnson's 250 D-viosterol in halibut liver oil, diluted in maize oil.) After 23 to 25 days the animals were sacrificed. The lead content of the blood was determined by the method of Willoughby, *et al.*[6] The lead content of the femora was determined by a modification of the above method (to be described).

The results of the experiments are presented in Table I. The vitamin D groups contained a higher percentage of lead in both the dried bone and the bone ash. Parallel effects were noted in the blood lead concentration of groups A and B. In group C which received the high phosphorus low calcium diet, the concentration of

[5] Bills, C. E., Honeywell, E. M., Wirick, A. M., and Nussmeier, M. J., *J. Biol. Chem.*, 1931, **90**, 619.

[6] Willoughby, C. E., Kraemer, E. O., and Smith, F. L., *Ind. Eng. Chem.*, Anal. Ed., 1935, **7**, 33.

TABLE I.
Influence of Dietary Calcium and Phosphorus upon Action of Vitamin D in Experimental Lead Poisoning.

	Group A		Group B		Group C	
	Diet A	Diet A + Vit. D	Diet B	Diet B + Vit. D	Diet C	Diet C + Vit. D
No. of animals	8	8	4	4	5	5
Avg. change in wt., g.	+3.3	+1.3	+21.0	+9.0	+12	+7
Avg. ash of fat free femora, %	28.3	34.7	26.5	40.4	40.0	35.7
Avg. Pb in fat free femora, mg. per 100 g.	160	326	73.3	216.8	115.3	305.2
Avg. Pb in bone ash, mg. per 100 g.	565	1230	291	542	292	778
Avg. Pb in whole blood, mg. per 100 cc.	0.71	1.3	0.22	0.68	>0.1	>0.1

lead in the blood was too low to be accurately determined. It is worthwhile to note, however, that even though this group received 3 times as much lead in the diet as the high calcium low phosphorus group, the blood lead concentrations in group C are distinctly below those of group B. The beneficial effects of the high phosphorus diet in experimental and clinical lead poisoning[2, 3] may be thus explained in terms of lowered blood lead concentration. Furthermore, in spite of the much lower blood lead concentration in group C as compared to group B, slightly more lead was deposited in the bones of the former group. To elucidate this apparent contradiction more knowledge is required of the thermodynamic activity of lead ions and phosphate ions in the blood stream. It is hoped that further investigations, which are in progress, will throw additional light upon the subject.