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THE UTILIZATION OF PHOSPHORUS FROM DIFFERENT SOURCES BY THE RAT.

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Godden and Ray (1938) commented on the conflicting statements in the literature with regard to the relative availability to animals of the phosphorus in supplements such as bone meal, di- and tricalcium phosphate. On the basis of balance experiments with sheep they came to the conclusion that the phosphorus of dicalcium phosphate was 50 per cent. more available than that of steamed bone flour. Later studies revealed that whilst most pyro- and metaphosphates were poorly utilized or completely unavailable, all orthophosphates such as bone meal, mono-, di-, and tricalcium phosphate were excellent sources of phosphorus in animal nutrition with little, if any, difference between them. (Barrentine *et al.*. 1944; Bird *et al.,* 1945; Ellis *et al.,* 1945; Gillis *et al.,* 1948; Louw, 1948; Cabell *et al.,* 1950; Reinach and Louw, 1950).

In view of the extreme importance of phosphate supplementation in this country coupled with the need for economical utilization of the limited supplies, it was decided that more information is needed concerning the relative efficiency of the available phosphatic products. Accordingly, further studies were undertaken and the results reported in this paper deal with the assimilation of phosphorus from the supplements bone meal, bone ash, dicalcium phosphate and tricalcium phosphate (c/f Table I).

Bone meal continues to be popular with the farming community. However, because of inadequate supplies and the fact that its manufacture requires special steam sterilizing plant not readily available on the ordinary farm, many farmers, especially in South West Africa, resorted, during the last world war, to preparing their own phosphatic supplement by incinerating bones in a specially constructed oven. It is of interest to recall many letters received at the time from farmers expressing a belief that much of the efficiency of the bone salts was lost in the process of burning.

The bone ash and bone meal used in this experiment were ground to pass a 70-mesh sieve, the former having been prepared from the latter by incineration in an electric muffle at a temperature of about 500° C. Di- and tricalcium phosphates are normally supplied in powder form and are for this reason less acceptable for the preparation of licks or for dosing purposes than the granular bone meal. However, they are suitable for incorporation into dairy or poultry meals and consequently were included in this study.

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TABLE 1.

Percentage Composition of Mineral Supplements.

	Calcium.	Phosphorus.
Bone Meal	21·36	9·86
Bone Ash	35·96	16·54
Dicalcium Phosphate, C.P.	23·49	18·13
Tricalcium Phosphate, Merck's extra pure	36·87	18·75

EXPERIMENTAL PROCEDURE.

The methods employed in evaluating the phosphates, including the use of a basal diet containing 0·05 per cent. P, were the same as those described previously by Reinach and Louw (loc. cit.). Only one trial was conducted to test the four materials. To this end ten quintets of young albino rats weighing approximately 50 grams at 35 days of age were selected. Rats within quintets were of the same age, sex and live weight. To arrive at values for the phosphorus content of the rats at the commencement of the experiment one animal from each quintet was killed and analysed. The four rats remaining in each quintet were distributed among the experimental diets described in table 2, the feed intake being equalized within quartos to that of the animal consuming the least. The experiment lasted 8 weeks. Growth, femur ash and total phosphorus retention were again used as criteria in deciding the relative efficiency of the several phosphatic products.

TABLE 2.

Composition of the Diets.

Ingredients.	Diets.			
	A.	B.	C.	D.
Egg White	150	150	150	150
Gelatine	30	30	30	30
Bacto Agar	20	20	20	20
Sucrose	40	40	40	40
Salt Mixture*	30	30	30	30
Harris Yeast	15	15	15	15
Butterfat	80	80	80	80
Cod Liver Oil	20	20	20	20
Dextrinized Starch	606·0	608·0	603·8	607·6
Dicalcium Phosphate	5·1	—	—	—
Tricalcium Phosphate	—	4·6	—	—
Bone Meal	—	—	9·3	—
Bone Ash	—	—	—	5·6
Calcium Carbonate	3·9	2·4	1·9	1·8
TOTAL	1000·0	1000·0	1000·0	1000·0
Percentage Ca	0·31	0·31	0·33	0·32
Percentage P	0·16	0·16	0·17	0·16

* Day and McCollum (1939), with $CaCO_3$ omitted.

Results and Discussion.

On account of the variations in the relative calcium and phosphorus contents of the supplements studied and the desirability for equalizing, by the addition of calcium carbonate, the ratios of these minerals in the rations fed (see table 2), it was, strictly speaking, not possible to make a critical comparison of the assimilation of both calcium and phosphorus in the same experiment. Data for calcium have, however, been included in table 3, giving a summary of the mean values for all treatments.

Diets A to D differed in respect of the source of their calcium and phosphorus contents only. Variations observed in the response of the four groups of animals on equalized feed intakes could, therefore, be ascribed only to differences in the availability of these elements in the test supplements. In this connection it should be mentioned that since the basal diet contained about 0·05 per cent. P the supplements furnished approximately 70 per cent. of the total phosphorus intake.

The rats on diet C (bone meal) apparently made less live weight gain than those on the other three diets. However, the greatest difference, that between diets A and C, just failed to have statistical significance. Tricalcium phosphate (diet B) lagged behind the other phosphate supplements in providing phosphorus for bone formation. This is apparent from an inspection of the figures for both the total and the percentage ash in the femurs of the rats, the differences in these respects between tricalcium phosphate and the supplement showing up best, viz., dicalcium phosphate (diet A) being significantly in favour of the latter (c.f. table 3).

The figures for percentage retention of the ingested bone-forming elements were based on the assumption that all the rats of a quintet contained initially equal amounts of calcium and phosphorus. These base values were obtained from an analysis of the animals sacrificed at the commencement of the experiment. Figures for percentage retention in respect of phosphorus calculated thus from the available data were 72·1, 65·9, 65·4 and 69·2 for dicalcium phosphate, tricalcium phosphate, bone meal and bone ash, respectively. The first-mentioned supplement was significantly better than the second and the third at $P = \cdot 01$. Bone ash proved to be superior to bone meal at $P = \cdot 05$. Somewhat similar differences were observed in the retention values for the calcium in the several diets.

The relatively low values observed for the percentage ash of the femurs (see table 3) indicated that the level of phosphate intake (0·16 per cent. P) at which the supplements were compared was well below that required for optimum bone nutrition. Under these circumstances, held to be favourable for bringing out differences in efficiency of utilization, the results could be summarized by saying that the phosphorus of dicalcium phosphate was assimilated better than that of any of the other products tested. Also, bone ash was found somewhat superior to the bone meal from which it was prepared by incineration.

It is, however, doubtful whether the observed differences in availability, shown to have statistical significance, are of importance also under practical conditions of feeding. In this connection the data in table 4, calculated from information in tables 1 and 3, are of interest.

For instance, 100 parts by weight of bone meal are equivalent to 59·6 parts by weight of bone ash on the basis of total phosphorus content, but to 56·2 parts of the latter product when availability to animals is taken into account. When replacing bone meal with certain of the other supplements on the basis of available instead of total phosphorus, a measurable saving could thus be effected. The practicability of this saving would, however, be dependent upon the method of utilizing

TABLE 3.

Treatment Averages.

	Treatments.				Necessary Difference.		Coefficient of Variance (%).
	A.	B.	C.	D.	P = ·05.	P = ·01.	
Feed consumed, gm.	290	290	290	290	—	—	—
Gain in Weight, gm.	71	70	67	69	4·03	5·44	6·3
Ash in dry, fat-free Femur, gm.	0·0841	0·0773	0·0806	0·0812	0·0034	0·0045	4·6
Ash in dry, fat-free Femur, Per Cent	51·0	49·9	51·3	51·2	1·064	1·437	2·6
Phosphorus.							
Total Intake, mg.	467·9	469·1	481·6	473·5	—	—	—
Initial Amount in Body, mg.	250·4	250·4	250·4	250·4	—	—	—
Final Amount in Body, mg.	589·6	561·5	567·9	577·1	—	—	—
Gain, mg.	339·2	311·1	317·5	326·7	—	—	—
Retention, Per Cent.	72·1	65·9	65·4	69·2	3·56	4·81	5·9
Calcium.							
Total Intake, mg.	909·1	912·2	950·0	937·7	—	—	—
Initial Amount in Body, mg.	335·8	335·8	335·8	335·8	—	—	—
Final Amount in Body, mg.	820·7	765·4	802·4	799·8	—	—	—
Gain, mg.	484·9	429·6	466·6	464·0	—	—	—
Retention Per Cent.	53·3	47·1	48·6	49·9	3·03	4·10	6·6

the supplement. The manufacturer of balanced rations, handling large quantities of materials at a time, would find the adjustment to an available basis feasible and, no doubt, advantageous. On the other hand to the farmer making use of the so-called "crush method" in which animals are given a small daily dose of a supplement, this adjustment would be impracticable. It should also be remembered that in practice, where supplements may be expected to be added in amounts above the minimum level of intake used in this investigation, small differences in efficiency may decrease or disappear entirely.

TABLE 4.

Comparative Values of the Supplements.

	Weight equivalence on the Basis of—	
	Total Phosphorus Content.	Available Phosphorus Content.
Bone Meal....................................	100	100
Bone Ash......................................	59·6	56·2
Dicalcium Phosphate............................	54·4	49·3
Tricalcium Phosphate...........................	52·6	52·1

It seems justified, therefore, to conclude that whilst the differences in availability to animals among certain supplements were such as to fulfil the demands for statistical significance they could for most practical purposes be ignored. In these circumstances the relative values of ortho-phosphatic supplements are determined by their content of total phosphorus only.

SUMMARY.

An experiment has been carried out in which the relative availability to young rats of the phosphorus in bone meal, bone ash, dicalcium phosphate and tricalcium phosphate has been ascertained. Live weight gains, the ash content of the femurs, and the total retention of phosphorus have been used as criteria in evaluating the supplements.

Dicalcium phosphate was found superior to the other three supplements. Bone ash proved to be on a par with, if not better than bone meal. Tricalcium phosphate was least efficient as a source of phosphorus for bone formation.

The practical significance of the observed differences in availability is briefly discussed.

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The practical significance of the observed differences in availability is briefly statistician, for carrying out the statistical analysis of the results and for suggestions in connection with their tabular presentation.

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