LXXXVI. THE INFLUENCE OF VARIATIONS IN THE SODIUM-POTASSIUM RATIO ON THE NITROGEN AND MINERAL METABOLISM OF THE GROWING PIG.

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THE experiments recorded in this paper were carried out as part of an investigation being conducted at this Institute on the mineral requirements of the growing pig. Certain results, not yet published, obtained by some of our co-workers engaged in this investigation, seemed to indicate that the addition of sodium to a ration of cereals led to increased rate of growth. The following metabolism experiments were conducted with the object of determining the influence of the addition of sodium salts to a cereal ration on the assimilation and retention of nitrogen and of those minerals which are most essential as constructive material for the growing pig.

PREVIOUS WORK.

Previous work on the effect of sodium and potassium salts in nutrition appears to have been almost entirely restricted to two questions, viz. (a) the effect of the addition of common salt in the feeding of farm animals, which forms one of the oldest subjects of agricultural research, and (b) the effect of increase of potassium salts on the excretion of sodium and chlorine, a question investigated by Bunge [1873], on the basis of whose one-day experiments the theory has been built up and generally accepted that intake of potassium salts leads to impoverishment of the organism in sodium chloride.

(a) *Effect of sodium chloride in animal feeding.* After various contradictory results recorded in the earlier part of the 19th century, Voit [1860] found that the addition of salt increased to a small extent the assimilation of nitrogenous substances, and this view was adopted for many years. Later researches, however, seem to show that its influence on nitrogen assimilation depends on the quantity in which it is taken. According to Belli [1904], who gives also a summary of the earlier literature on the subject, in the small amounts which are naturally present in foodstuffs it has a favourable influence, while in the quantities in which it is usually added to the food the result is unfavourable.

(b) *The effect of increase of potassium salts.* In his experiments on himself Bunge found that by taking doses of potassium in the form of the phosphate, the citrate or the sulphate, there was a very large increase in the excretion of sodium and chlorine in the urine. In a converse experiment, by taking a dose of sodium citrate, he found similarly an increase in the urinary excretion of potassium, the excretion of chlorine on the other hand showing a slight diminution. Two points are specially to be noted in regard to Bunge's work: (1) he analysed only the urine, no attention being paid to faecal excretion, and (2) his own figures show that the excessive loss of sodium and chlorine on the day of the experiment was made up for on the three days following the potassium intake, when the excretion of these elements was far below the normal. It is further noteworthy that Bunge himself, in summarising his work and mentioning future experiments that must be carried out, refers specially to the necessity for analysing the intestinal excretions, and for determining whether repeated intake of potassium salts would lead to continued excessive excretion of sodium and chlorine.

In spite of these admissions, and of the limited nature of his own experiments, Bunge maintains his point that the organism is impoverished in sodium chloride by the intake of potassium salts, and he proceeds to elaborate a theory that the craving for salt experienced by herbivorous animals and peoples [Bunge, 1874] is due to the large amount of potassium salts contained in vegetable foodstuffs, the excess of potassium leading to excessive excretion of sodium chloride, which has to be made good by increased intake of salt with the food, while on the other hand carnivorous animals shew no such craving.

In this connection it is perhaps worthy of mention that Köppe [1900] in discussing the salt craving shown by herbivorous animals, points out the difference between the ash-content and the salt-content of plants. He states that, in spite of their high ash-content, plants contain only a very small amount of dissolved and dissociated inorganic salts. The salts present are mostly in organic combination, while animal fluids on the other hand have a high content of free inorganic salts. This may also have a bearing on the fact that herbivorous animals, such as cattle and sheep, may be fed for a long time on potassium-rich food without any addition of salt, and show no visible disturbance in their development and condition. As Abderhalden [1923] points out, it is quite possible and indeed probable that the potassium salts of foodstuffs exercise no such strong influence as is brought about by the introduction into the body of potassium salts in large quantities in the inorganic form.

H. G. Miller [1923] investigated the influence of potassium on urinary sodium and chlorine excretion, his paper being published after the present series of experiments had been begun. His object was to find the effect of *continued* large additions of potassium to the diet, his experiments being carried out on a pig, but, like Bunge, he analysed only the urine, so that his results can lead to no conclusions as to the impoverishment of the organism

in sodium chloride. But, as a matter of fact, he finds that even in the urine alone, the increased sodium excretion is only transitory, in spite of continued administration of potassium.

EFFECT OF SODIUM AND POTASSIUM SALTS ON CALCIUM ASSIMILATION.

Bunge's theory was taken up by Seemann [1879] who believed that he had found in it the cause of rickets. He argues thus: excessive supply of potassium causes excessive excretion of chlorine and hence want of chlorides in the body; this leads to insufficient formation of hydrochloric acid in the stomach, and owing to this deficiency of free acid the calcium salts of the food are not sufficiently dissolved in the stomach, thus leading to rickets. Zander [1881] followed up Seemann's work by investigating cases of rickets in breast-fed children. He found that in the case of healthy children the mother's milk contained potassium and sodium in the proportion of 2 : 1, while in the case of rachitic children the ratio was $3-4\frac{1}{2}$: 1. Hence Zander considers Seemann's theory fully confirmed.

The only other reference found to the effect of sodium and potassium salts on calcium assimilation is that of Aron [1905], who states: "From researches undertaken to investigate licking disease in cattle, it appears very probable that the content of the food in potassium and sodium influences the deposit of calcium and the bone formation, viz., with strongly diminished sodium content of the food, and at the same time very high potassium content, the deposit of calcium, and also the bone growth, remains below normal, in spite of abundant supply of calcium and phosphorus." No details are given by Aron, and the later paper which he promises on the subject seems never to have been published.

PRESENT INVESTIGATION.

While the primary object of the present investigation was to ascertain the effect on calcium assimilation of altering the sodium-potassium ratio in an animal's diet, it seemed advisable also to study the effect on the general metabolism, and at the same time to find whether Bunge's conclusions were justified.

Methods.

The experiments were carried out on young pigs by the methods described in a previous paper by the present authors on the influence of oils on the assimilation of calcium and phosphorus in the growing pig [Husband, Godden and Richards, 1923] but in addition to determining the total nitrogen, calcium and phosphorus in the urine and faeces, there were also determined magnesium, chlorine, sodium and potassium; magnesium by the volumetric method of Tisdall and Kramer [1921]; chlorine by the Volhard method; potassium by the cobaltinitrite method of Green [1912]; sodium and potassium as mixed sulphates, and hence sodium by difference.

Experimental Data.

Basal diet. The basal diet used in the two experiments to be described consisted of maize, oatmeal, barley meal and blood meal in the proportions stated below. Analyses of the foodstuffs gave the following figures (averages of the samples used in the two experiments):

Average composition of the basal diet.

Proportions		Total N %	CaO %	P ₂ O ₅ %	MgO %	Cl %	Na ₂ O %	K ₂ O %
Maize	10 parts	1.46	0.007	0.645	0.193	Nil	0.040	0.500
Oatmeal	10 "	2.19	0.082	1.068	0.232	Nil	0.067	0.618
Barley meal	10 "	1.22	0.056	0.718	0.164	0.007	0.061	0.659
Blood meal	1 part	13.37	0.141	0.301	0.037	0.934	1.196	0.414

The exact intake of nitrogen, CaO and P₂O₅, etc., in each experiment is stated in the tables giving the experimental data.

The amount of CaO in the ration was increased in each experiment by the addition of 25 cc. of a 20 % solution of calcium chloride, and in Exp. 2 it was still further increased by adding 6 grams of chalk. In addition the animal received 10 cc. of olive oil daily, the previous work having shown that this assisted regular defaecation, and helped to keep the animal in good condition. Any effect of the olive oil on assimilation would be continuous throughout the experiment, and may therefore be excluded in considering the results.

Exp. 1. To determine the effect of the addition of sodium chloride to the ration. In this experiment, which extended over 19 days, the animal received 775 g. of the basal ration plus 25 cc. of 20 % calcium chloride solution per day. From the 6th to the 12th day 50 cc. of a 10 % solution of sodium chloride were added to the daily ration, this being withdrawn again as from the 13th day. The results of the analyses are given in Table I and shown in part in Fig. 1. In order to economise space, only the averages for the periods are given in the tables, but it may be mentioned that the variations in the day to day figures are small.

It will be seen that the results of the administration of sodium chloride point to a definite increase in the rate of retention of the chief constructive materials required for proper growth. The balances for N, P₂O₅ and CaO, which had been falling during the preliminary period, all showed an increase during the experimental period. The results for MgO, given in the tables, are not meanwhile discussed, but it may be stated that, in general, the curves obtained resemble those for phosphorus.

With regard to the excretion of K₂O, the interesting result is noted that, while the feeding of sodium chloride caused a large increase in the urinary excretion of K₂O, as observed by Bunge, this was accompanied by a large decrease in the faecal excretion, so that the balance for the experimental period remained practically the same as in the pre-period, and in the post-period showed a distinct increase (see Fig. 1). This point will be referred to again in the discussion of the results after Exp. 2.

Sodium chloride experiment. Duration of experiment: 19 days.

Days	Average daily wt. of faeces	Nitrogen					CaO				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-5 inclusive	329	15.15	6.74	3.62	10.36	4.79	1.74	0.07	1.56	1.63	0.11
→ 6-12 "	289	"	6.71	3.27	9.98	5.17	"	0.08	1.23	1.31	0.43
← 13-19 "	276	"	6.96	3.20	10.16	4.99	"	0.06	1.12	1.18	0.56
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Days	Average daily wt. of faeces	P ₂ O ₅					MgO				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-5 "		5.94	1.28	3.63	4.91	1.03	1.49	0.06	1.34	1.40	0.09
→ 6-12 "		"	1.62	3.22	4.84	1.10	"	0.06	1.29	1.35	0.14
← 13-19 "		"	1.51	3.22	4.73	1.21	"	0.05	1.26	1.31	0.18
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Days	Average daily wt. of faeces	Cl					Na ₂ O				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-5 "		1.97	2.50	0.10	2.60	-0.63	0.53	0.31	0.05	0.36	0.17
→ 6-12 "		4.84	4.82	0.12	4.94	-0.10	3.03	2.32	0.20	2.52	0.51
← 13-19 "		1.97	2.20	0.11	2.31	-0.34	0.53	0.45	0.11	0.56	-0.03
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Days	Average daily wt. of faeces	K ₂ O									
		Intake	Urine	Faeces	Total	Balance					
1-5 "		4.38	0.59	2.47	3.06	1.32					
→ 6-12 "		"	1.31	1.83	3.14	1.24					
← 13-19 "		"	0.83	2.06	2.89	1.49					

→ indicates the introduction of sodium chloride.

← indicates the withdrawal of sodium chloride.

Fig. 1. K_2O excretions and balances. Experiment 1, Table I.

Exp. 2. To show the effect of the addition of sodium citrate to the ration.

As there was a possibility that the improved balances in the experiment just recorded might have been due to the chlorine and not to the sodium of the salt administered, in this experiment sodium citrate was added to the

basal ration instead of sodium chloride. As before, the animal received daily 775 g. of the basal ration, with 25 cc. of 20 % calcium chloride solution and 6 g. of chalk. In the experimental period it received in addition, per day, 50 cc. of a solution of sodium citrate containing approximately the same amount of Na_2O as the sodium chloride solution of Exp. 1. In this experiment the periods were lengthened—preliminary period 16 days, the results being given for the last eight days of the period; experimental and post periods 14 days each. The results of this experiment are given in Table II and Figs. 2 and 3, and it will be seen that they confirm those of the first experiment.

Table II. *Exp. 2. Average daily excretions and balances in grams.*

Sodium citrate experiment. Duration of experiment: 36 days.											
Days (inclusive)	Av. daily weight of faeces	Nitrogen					CaO				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-4	260	15.88	6.27	3.87	10.14	5.74	5.26	0.39	2.56	2.95	2.31
5-8	282	"	6.26	3.93	10.19	5.69	"	0.45	2.60	3.05	2.21
→ 9-15	281	"	6.05	3.87	9.92	5.96	"	0.46	2.43	2.89	2.37
16-22	252	"	6.06	3.38	9.44	6.44	"	0.68	2.00	2.68	2.58
← 23-29	216	"	6.22	3.03	9.25	6.63	"	0.65	1.80	2.45	2.81
30-36	202	"	5.95	3.12	9.07	6.81	"	0.48	1.83	2.31	2.95
		P_2O_5					MgO				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-4	—	6.36	0.27	3.42	3.69	2.67	1.48	0.11	1.23	1.34	0.14
5-8	—	"	0.29	3.58	3.87	2.49	"	0.11	1.27	1.38	0.10
→ 9-15	—	"	0.30	3.61	3.91	2.45	"	0.09	1.32	1.41	0.07
16-22	—	"	0.29	3.19	3.48	2.88	"	0.14	1.24	1.38	0.10
← 23-29	—	"	0.23	2.93	3.16	3.20	"	0.12	1.19	1.31	0.17
30-36	—	"	0.22	2.94	3.16	3.20	"	0.13	1.21	1.34	0.14
		Cl					Na_2O				
		Intake	Urine	Faeces	Total	Balance	Intake	Urine	Faeces	Total	Balance
1-4	—	2.34	2.54	0.18	2.72	-0.38	0.91	0.52	0.49	1.01	-0.10
5-8	—	"	2.51	0.20	2.71	-0.37	"	0.42	0.49	0.91	+0.0
→ 9-15	—	"	2.56	0.19	2.75	-0.41	3.44	1.79	1.11	2.90	+0.54
16-22	—	"	2.49	0.18	2.67	-0.33	"	2.19	1.17	3.36	+0.08
← 23-29	—	"	2.56	0.15	2.71	-0.37	0.91	0.70	0.61	1.31	-0.40
30-36	—	"	2.52	0.17	2.69	-0.35	"	0.48	0.50	0.98	-0.07
		K_2O									
		Intake	Urine	Faeces	Total	Balance					
1-4	—	4.71	1.42	1.53	2.95	1.76					
5-8	—	"	1.34	1.74	3.08	1.63					
→ 9-15	—	"	2.26	1.24	3.50	1.21					
16-22	—	"	2.05	1.20	3.25	1.46					
← 23-29	—	"	1.35	1.40	2.75	1.96					
30-36	—	"	1.36	1.52	2.88	1.83					

→ indicates the introduction of sodium citrate.

← indicates the withdrawal of sodium citrate.

Nitrogen, calcium and phosphorus assimilation. The balances for N, CaO and P_2O_5 in both experiments all show an increase during the experimental period, the increases continuing into the post-period, except that for N in Exp. 1, which fell back slightly. In Exp. 2, where the longer periods enable the results to be followed more closely (see Fig. 2), it will be seen that the improvements are all more marked during the second week of the sodium feeding, and that the increases fall off gradually in the post-period.

Urinary excretion of CaO. A noteworthy point in Exp. 2 was the very marked effect of the sodium citrate on the urinary excretion of CaO, which

Fig. 2. N, CaO, and P_2O_5 balances for Experiment 2, Table II. Fig. 3. K_2O excretions and balances. Experiment 2, Table II.

increased steadily during the experimental period to about twice the original amount, and in the post-period fell steadily back again. The following figures give the urinary excretion of CaO in grams per successive two day periods:

Pre-period	...	...	—	—	—	0.70	0.85	0.87	0.90
Sodium citrate period	0.76	0.85	0.97	1.30	1.31	1.36	1.43		
Post-period	...	...	1.52	1.44	1.12	0.98	1.05	0.95	0.83

Taking the increased urinary excretion in conjunction with the increased CaO balance, this seems to point clearly to increased assimilation of CaO consequent on the addition of sodium salts to the diet.

Chlorine balance. Throughout both the experiments the chlorine balance remained negative. The addition of the sodium citrate seemed to have no lasting effect, a small positive balance for the first two days of the experimental period being counterbalanced by a large negative balance on the two following days. The average balance for the periods remained practically the same during the whole experiment. In Exp. 1, the addition of sodium chloride brought the animal nearly to equilibrium as regards chlorine. Of the additional chlorine fed practically the whole amount was excreted in the urine, the faecal excretion increasing only very slightly. On the other hand, the faecal excretion of Na_2O increased quite noticeably during the sodium chloride period, though the urinary increase was of course much greater. This would indicate that, of the additional sodium chloride fed, the animal assimilated more of the chlorine than of the sodium, in accordance with the fact that the chlorine balance was negative whereas that for sodium was positive.

Na_2O balance. In Exp. 1 the balance increased during the experimental period, and in the post-period went back to slightly less than in the pre-period. In Exp. 2 the large increase in the Na_2O balance for the first week of the sodium citrate period almost disappeared during the second week. Further, this positive balance was neutralised by a large negative balance which followed the withdrawal of the sodium citrate and was itself also only temporary.

K_2O balance. One of the most important points brought out by these experiments, and shown very clearly in Figs. 1 and 3, is to be found in a consideration of the K_2O results. Attention has already been drawn to the fact that in Exp. 1 the increased urinary excretion of K_2O was compensated for by the diminished faecal excretion (see Fig. 1); Exp. 2 confirmed this result. The excessive excretion of K_2O in the urine was largely counterbalanced by diminished excretion of K_2O in the faeces, and though the balance for the experimental period was below that of the pre-period, Fig. 3 shows that, for the second week, the balance was adjusting itself towards the former level, and withdrawal of the sodium citrate brought it immediately well above the original figure. From the analytical data we find that in Exp. 1 the total excretion of K_2O for the whole period of the experiment was 57.59 g. while the total excretion for the period, based on the average for the preliminary period, would have been 58.23 g. Taking the urinary excretion of K_2O , the amount actually excreted during the experiment was 17.98 g., while the amount based on the average of the preliminary period would have been 11.29 g. That is, while there was an increase of nearly 60 % in the urinary excretion of K_2O , the total amount of K_2O excreted by the organism was actually slightly less than normal. The figures for Exp. 2 show similar results. The total excretion of K_2O for the 36 days of the experiment was 110.59 g. as against 108.4 g. the amount calculated from the average of the preliminary period, *i.e.* the amount excreted was only 2 % above normal. The urinary excretion for the same period was 60.05 g. as against the calculated figure 49.54 g., *i.e.* the urinary excretion was 21 % above the normal.

This proves conclusively that urinary figures alone form no guide as to the balance of the organism in Na_2O and K_2O when the ratio of these is altered in the diet. Thus one of the points suggested by Bunge as necessary in future investigation, viz. analysis of the faecal excretion, turns out to be of such vital importance that quite erroneous conclusions have been drawn from the incomplete data of his experiments. Another of the points he brought forward, viz. the effect of continued administration of potassium, has been investigated by Miller [1923], who found in his experiments that the excessive excretion of sodium and chlorine in the urine was only transitory, little effect being observed after the first twenty-four hours period, even on continuing the doses of the potassium salt in the diet. This is not in accordance with our findings in the converse experiment. While there are indications in the figures for the experimental period that the animal is gradually adjusting itself to the excess of sodium salt in the ration, nevertheless after 14 days' feeding of sodium citrate, the amount of K_2O in the urine remains very much higher than in the pre-period, and withdrawal of the sodium salt is followed by an immediate drop in the urinary potassium excretion. Thus the effect in our investigation is by no means so transitory as Miller has found in his potassium feeding experiments.

It remains, therefore, still a moot point whether the organism would adjust itself so completely to the change of diet that the urinary excretion of K_2O would return to the original figure, but in view of our findings this point seems to us of minor importance. Of much more significance is the fact that the organism has apparently the power of adjusting the balance so that, even while abnormal urinary excretion continues, the balance may be very little disturbed, and when disturbed has the tendency to come back to normal.

The importance of a proper physiological balance between the mineral constituents of a ration has already been emphasised by Orr [1922, 1923], and further investigation is proceeding along these lines with special reference to the sodium-potassium ratio.

CONCLUSIONS.

1. The addition of sodium chloride or sodium citrate to a ration of cereal grain fed to a growing pig led to increased assimilation and retention of nitrogen, calcium and phosphorus.
2. The increase of sodium salts in the ration caused increased excretion of potassium in the urine, the increase being still very marked even after 14 days' continued administration of the sodium salt.
3. The excessive urinary excretion of potassium was more or less completely counterbalanced by diminished faecal excretion, with the result that the balance showed very little deviation from the normal, even during a period of very abnormal urinary excretion. Hence metabolic experiments must include analyses of both faeces and urine, otherwise the conclusions drawn may be completely erroneous.

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