CLINICAL CALORIMETRY.

XLVI. PROLONGED MEAT DIETS WITH A STUDY OF THE METABOLISM OF NITROGEN, CALCIUM, AND PHOSPHORUS.

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Analysis of Meat.

Two arctic explorers lived for 1 year on a diet consisting exclusively of meat, as described in a previous communication (1). In order to analyze their diet, we duplicated, as nearly as possible, the food taken by them while they were in the metabolism ward. The meat consumed during the earlier periods of observation was from animals killed the preceding day, but after the 3rd month, ordinary refrigerated meat was used. The samples were put into closed but not air-tight containers, and kept in an ice box for periods of 3 to 10 days. Analyses were then made of aliquot portions of the collected meat samples for each period. Determinations of nitrogen and fat were made on twenty individual samples of muscle and six of tongue.

The lean meat was weighed, ground in a meat grinder, and an aliquot portion dried on a steam bath. The dried material was weighed, passed through a grinder to break up the lumps, and stored in Mason jars. It was further pulverized in a mortar before samples were taken for analysis. For nitrogen determinations, duplicate samples of 0.5 gm. each of the dried material were used. These were digested with a mixture consisting of 50 cc. of 5 per cent $CuSO_4$ solution, 300 cc. of 85 per cent phosphoric acid, and 100 cc. of concentrated sulfuric acid. The Kjeldahl method was followed. Pumice was added to reduce bumping. Fat determi-

TABLE I.

Analyses of Meat: Nitrogen and Fat.

Gm. per 100 gm. wet weight.

Nature of meat.	No. of samples.	Nitrogen.			Fat.		
		Maximum.	Minimum.	Average.	Maximum.	Minimum.	Average.
Beef muscle (well trimmed).	27	3.77	2.78	3.24	7.80	2.50	5.02
" " (not ").	9	3.60	2.84	3.18	20.80	7.70	11.78
" tongue	17	3.65	2.70	3.15	34.60	14.70	24.24
" liver	14	3.34	2.61	2.97	8.55	3.58	5.85
" kidney	2	2.47	2.23	2.35	2.80	2.40	2.60
" brain	1			1.68			9.30
" fat	18	0.34	0.17	0.22	92.70	86.00	90.20
" bone marrow	8	0.25	0.15	0.20	90.30	87.80	89.10
Veal	2	3.00	2.76	2.88	17.21	13.40	15.31
Lamb	4	2.56	2.23	2.42	33.00	22.90	26.85
Bacon	5	1.40	1.08	1.23	63.00	57.50	59.50

nations were made on 5.0 gm. portions of the dried meat by extracting them overnight in a Soxhlet apparatus with ethyl ether and purifying the extract with petroleum ether.

Samples of fat and marrow were passed through a meat grinder, three or four times, and aliquot portions were preserved in the ice box. The true fat content of these samples was determined as follows: Approximately 10 gm. of material were weighed in a Petri dish and dried on a steam bath to constant weight, thus the water content of the fat was obtained. Warm gasoline was added to dissolve the fat, and the non-soluble matter was separated by use of a Gooch crucible. The residue in the crucible was washed several times with warm gasoline and finally with ether. The crucible was dried to constant weight and the non-soluble matter weighed. After the weight of the water and of the non-soluble ingredients was deducted from the original weight of the sample, the remainder was taken as the true fat content. The material which was insoluble in ether was collected from a number of samples and analyzed for its nitrogen content which was found to be close to 10 per cent. This figure was used in calculating the nitrogen content of the fat after determining the residue which was insoluble in ether. The same procedure was adopted with the marrow.

TABLE II.

Analyses of Meat: Calcium and Phosphorus.

Gm. per 100 gm. wet weight.

Nature of meat.	No. of samples.	Calcium.			Phosphorus.		
		Maximum.	Minimum.	Average.	Maximum.	Minimum.	Average.
Beef muscle	17	0.0180	0.0084	0.0118	0.260	0.194	0.223
" tongue	9	0.0091	0.0063	0.0075	0.214	0.181	0.201
" liver	13	0.0120	0.0058	0.0079	0.480	0.310	0.416
" kidney	1			0.0074			0.252
" brain	1			0.0076			0.380
" fat	5	0.0190	0.0100	0.0138	0.065	0.050	0.056
Veal	2	0.0146	0.0134	0.0140	0.216	0.191	0.204
Lamb	4	0.0330	0.0109	0.0210	0.190	0.170	0.181
Bacon	5	0.0187	0.0096	0.0132	0.120	0.090	0.108

Phosphorus was determined in samples of 1.0 gm. each by the volumetric molybdate method after digestion with nitric and sulfuric acids (2). Calcium was determined by McCrudden's method (3) on 10 gm. of dried material which had been ashed.

Table I contains the maximum, minimum, and average values for nitrogen and fat. The data for calcium and phosphorus are given in Table II. A large variation was found in the fat content of different samples of tongue, according to the portion of the tongue used, the base having the highest fat content. The muscle samples in the first group were very lean as all free fat was carefully trimmed off in the ward kitchen; those in the second group had a higher fat content because they were not trimmed. There was considerable variation in the amount of calcium in the different samples of the same kind of meat. This agrees with the findings of Heubner and Rona (4) and of Denis and Corley (5). The calcium content of meat as given by Sherman and Gettler (6) is 0.055 gm. of calcium per 100 gm. of protein. The average result of the seventeen determinations made by us on beef muscle was 0.059 gm. of calcium per 100 gm. of protein.

Absorption of Meat.

The absorption of food materials from the intestinal tract is an important feature in any study of nutrition. The unabsorbed portions play no part in the foodstuffs actually metabolized in the body. Prausnitz (7) found little variation in the amount of nitrogen in the feces with wide fluctuations in the amount of protein in the food. Bloor and his associates (8, 9) noted the presence of fat in the feces of animals on fat-free diets and emphasized the fact that such fat may come from sources other than the food ingested.

The feces were collected for periods of 3 to 10 days, preserved with acid-alcohol, and dried, and the nitrogen and fat determinations were made by the same methods as those used for food. No analyses for the amount of carbohydrate in the feces were made because the food contained only that carbohydrate which was present as glycogen in the meat. Identification of the nature of the nitrogen and fat found in the feces was not attempted. The significant data obtained are presented in Table III.

In the case of the subject K. A., the nitrogen loss in the feces showed a variation of 2.4 to 7.1 per cent with an average of 4.5 per cent during the periods when he was receiving meat alone. The subject V. S. lost from 6.5 to 10.4 per cent of his ingested nitrogen, an average of 7.5 per cent. A comparison with the control periods shows a better absorption of protein in K. A. while on the meat diet but no significant difference in V. S.

The loss of fat in K. A. varied between 1.7 and 4.8 per cent with an average of 3.0 per cent; the loss for V. S. ranged from 5.4 to 14.6 per cent with the average at 9.2 per cent. The absorption of fat in both subjects was essentially the same as during the control periods.

In the case of each subject, the percentage loss of both nitrogen

TABLE III.

*Percentage of Ingested Nitrogen and Fat Excreted in the Feces.**

Subject.	Period No.	No. of days.	Nitrogen.			Fat.			
			Food.	Feces.		Food.	Feces.		
			gm.	*gm.*	*per cent*	*gm.*	*gm.*	*per cent*	
K. A.	1	8	79.1	10.1	12.8	980	21.4	2.2	
	2	10	221.0	7.2	3.3	2315	44.6	1.9	
	3	10	198.5	10.7	5.4	2022	96.3	4.8	
	4	10	202.1	4.9	2.4	2527	43.6	1.7	
	5	10	185.8	13.1	7.1	2812	113.2	4.0	
	6	10	188.7	7.6	4.0	2438	59.5	2.4	
	7	10	196.1	9.5	4.8	2238	78.8	3.5	
	8	10	183.0	8.6	4.7	2158	67.1	3.1	
	9	10	203.1	7.7	3.8	2165	67.1	3.1	
	10	10	175.9	9.0	5.1	1623	41.7	2.6	
									Interval 8 mos.
	11	10	213.6	9.8	4.6	2082	60.0	2.9	
	12	10	216.3	8.7	4.0	1981	53.5	2.7	
	13	7	51.3	6.0	11.7	1593	103.0	6.5	
	14	7	50.4	5.7	11.3	1634	39.5	2.4	
	15	7	115.5	6.4	5.5	1447	33.0	2.3	
	16	12	96.8	17.6†	18.2	1337			
	17	12	174.5	14.3†	8.2	2434			
	18	10	125.0	13.3	10.7	1446	24.3	1.7	
Average per day, Periods 2–12.‡.....			19.9	0.9	4.5	221.5	6.6	3.0	
V. S.	1	5	64.0	5.4	8.4	750	82.5	11.0	
	2	4	115.9	7.7	6.6	609	65.0	10.7	
	3	9	146.2	9.8	6.7	1697	145.8	8.6	
	4	9	123.3	12.8	10.4	1044	152.2	14.6	
	5	10§							
	6	10§							
	7	10	236.8	17.2	7.3	2175	202.0	9.3	
									Interval 10 mos.
	8	8	127.3	8.3	6.5	1507	81.0	5.4	
	9	7	83.4	7.6	9.1	1652	21.8	1.3	
	10	7	89.8	12.6	14.0	1036	23.0	2.2	
Average per day, Periods 2–4, 7, and 8.‡...........			18.7	1.4	7.5	175.8	16.2	9.2	

* The explanatory remarks in Table IV, apply to periods as given in this table.

† Estimated as 10 per cent of the urinary nitrogen as found when on mixed diet in Periods 1 and 18.

‡ Exclusive meat diet.

§ Data for these periods are incomplete.

and fat remained nearly the same throughout the different periods. There was a wide difference, however, between the results in the

TABLE IV.

Nitrogen Balance.

Subject.	Period No.	No. of days.	Total intake.	Output. Urine	Output. Feces.	Output. Total.	Balance for period.	Remarks.
			gm.	*gm.*	*gm.*	*gm.*	*gm.*	
K. A.	1	8	79.1	65.3	10.1	75.4	+3.7	Mixed diet.
	2	10	221.0	202.6	7.2	209.8	+11.2	Meat.
	3	10	198.5	190.6	10.7	201.3	−2.8	"
	4	10	202.1	179.4	4.9	184.3	+17.8	"
	5	10	185.8	175.6	13.1	188.7	−2.9	"
	6	10	188.7	167.6	7.6	175.2	+13.5	"
	7	10	196.1	178.3	9.5	187.8	+8.3	"
	8	10	183.0	181.0	8.6	189.6	−6.6	"
	9	10	203.1	193.4	7.7	201.1	+2.0	"
	10	10	175.9	183.6	9.0	192.6	−16.7	"
	11	10	213.6	222.1	9.8	231.9	−18.3	"
	12	10	216.3	217.0	8.7	225.7	−9.4	"
	13	7	51.3	76.5	6.0	82.5	−31.2	Low protein, high fat.
	14	7	50.4	57.7	5.7	63.4	−13.0	" " " " 35 gm. carbohydrate.
	15	7	115.5	117.4	6.4	123.8	−8.3	Meat.
	16	12	96.8	176.2	17.6*	193.8	−97.0	Mixed diet, pneumonia.
	17	12	174.5	142.5	14.3*	156.8	+17.7	" " convalescent.
	18	10	125.0	92.3	13.3	105.6	+19.4	Mixed diet.
V. S.	1	5	64.0	54.2	5.4	59.6	+4.4	Mixed diet.
	2	4	115.9	106.2	7.7	113.9	+2.0	Meat.
	3	9	146.2	159.2	9.8	169.0	−22.8	"
	4	9	123.3	145.7	12.8	158.5	−35.2	"
	5	10						"
	6	10						"
	7	10	236.8	221.6	17.2	238.8	−2.0	"
	8	8	127.3	145.6	8.3	153.9	−26.6	"
	9	7	83.4	77.2	7.6	84.8	−1.4	Low protein, high fat, 35 gm. carbohydrate.
	10	7	89.8	63.1	12.6	75.7	+14.1	Mixed diet.

* Estimated as 10 per cent of the urinary nitrogen.

cases of the two individuals, although both were within normal limits. In each case, both nitrogen and fat were well absorbed.

Metabolism of Nitrogen, Calcium, and Phosphorus.

A meat diet, unless supplemented by some material, like bone, which is rich in calcium, is unsatisfactory from the mineral stand-

TABLE V.

*Calcium Balances.**

Subject.	Period No.	No. of days.	Total intake.	Output.			Balance for period.
				Urine.	Feces.	Total.	
			gm.	*gm.*	*gm.*	*gm.*	*gm.*
K. A.	1	8	2.950	1.784	1.360	3.144	−0.194
	2	10	0.611	3.131	1.088	4.219	−3.608
	3	10	0.555	4.107	1.645	5.752	−5.197
	4	10	1.540	3.978	0.950	4.928	−3.388
	5	10	1.346	3.060	2.420	5.480	−4.134
	6	10	0.975	3.252	1.108	4.360	−3.385
	7	10	1.011	3.012	1.192	4.204	−3.193
	8	10	1.034	3.360	1.147	4.507	−3.473
	9	10	0.713	3.190	1.088	4.278	−3.565
	10	10	0.932	2.842	0.855	3.697	−2.765
	11	10	1.167	2.550	0.977	3.527	−2.360
	12	10	1.178	2.300	0.631	2.931	−1.753
	13	7	0.789	1.932	1.000	2.932	−2.143
	14	7	2.420	3.440	1.290	4.730	−2.310
	15	7	0.637	1.770	0.590	2.360	−1.723
	16	12		3.800			
	17	12		3.800			
	18	10	5.826	3.080	3.540	6.620	−0.794
V. S.	1	5	2.172	0.339	1.158	1.497	+0.675
	2	4	0.496	0.392	1.148	1.540	−1.044
	3	9	0.716	1.228	1.588	2.816	−2.100
	4	9	0.625	1.069	2.015	3.084	−2.459
	5	10					
	6	10					
	7	10	1.166	1.470	7.910	9.380	−8.214
	8	8	0.644	0.784	1.320	2.104	−1.460
	9	7	2.451	1.360	1.083	2.443	+0.008
	10	7	3.431	0.789	2.194	2.983	+0.448

* The explanatory remarks as given in Table IV apply in this table.

point because of its low calcium content. Sherman (10) states that the normal calcium requirement of a man weighing 70 kilos is approximately 0.45 gm. per day. The calcium content of meat as given by Sherman and Gettler (6) is 0.055 gm. of calcium per 100 gm. of protein. Therefore it would require about 800 gm. of protein per day to supply the needed calcium.

TABLE VI.

*Phosphorus Balance.**

Subject.	Period No.	No. of days.	Total intake.	Output.			Balance for period.
				Urine.	Feces.	Total.	
			gm.	*gm.*	*gm.*	*gm.*	*gm.*
K. A.	1	8	6.59	3.88	1.68	5.56	+1.03
	2	10	13.61	14.21	1.07	15.28	−1.67
	3	10	13.04	12.58	1.39	13.97	−0.93
	4	10	13.39	12.65	0.76	13.40	−0.02
	5	10	13.75	11.52	2.05	13.57	+0.18
	6	10	15.09	13.11	1.28	14.39	+0.70
	7	10	17.01	14.76	1.53	16.29	+0.72
	8	10	14.76	12.88	1.25	14.13	+0.63
	9	10	17.12	13.68	1.12	14.80	+2.32
	10	10	14.90	12.96	1.48	14.44	+0.46
	11	10	19.19	16.72	1.74	18.46	+0.73
	12	10	18.45	17.56	1.68	19.24	−0.79
	13	7	5.21	6.89	1.12	8.01	−2.80
	14	7	5.16	5.21	1.29	6.50	−1.34
	15	7	10.61	10.12	1.39	11.51	−0.90
	16	12		13.01			
	17	12		13.01			
	18	10	11.81	9.02	3.49	12.51	−0.70
V. S.	1	5	4.89	3.00	1.71	4.71	+0.18
	2	4	8.65	5.84	2.57	8.41	+0.24
	3	9	15.67	11.50	2.82	14.32	+1.36
	4	9	8.18	9.45	3.15	12.60	−4.42
	5	10					
	6	10					
	7	10	21.49	15.51	6.90	22.41	−0.92
	8	8	10.63	10.16	2.07	12.23	−1.60
	9	7	6.69	6.96	1.49	8.45	−1.76
	10	7	8.37	5.10	3.43	8.53	−0.16

* The explanatory remarks as given in Table IV apply in this table.

A meat diet contains an excess of the acid-forming elements, sulfur and phosphorus. The effect of an acid diet on the excretion of calcium has been reviewed by Stewart and Percival (11). More recently Bauer, Albright, and Aub (12) have reported that an acid diet increases the calcium excretion and that the increase occurs principally in the urine.

The phosphorus metabolism with acid diets has not received as much consideration as the calcium metabolism. Fitz, Alsberg, and Henderson (13) gave rabbits a diet to which hydrochloric acid had been added and noticed first an increase and then a decrease in the output of phosphorus in the urine. Goto (14) noted an increase in the excretion of phosphorus in the urine of rabbits after the ingestion of acid-forming food.

The balances for the men which we studied are given in Tables IV to VI which contain the total intake and output for each period. The intake values for the meat periods were obtained by analysis as described in the first section of this paper; for the other periods, they were calculated from the tables prepared by Rose (15).

Nitrogen Balance.—While on the meat diet, K. A. was in nitrogen equilibrium. In Period 13, his appetite was poor, and he swung to the negative side, and in Period 16, when he had pneumonia, his balance was −8.0 gm. of nitrogen per day. V. S. had a definite negative balance in Periods 3, 4, and 8, while taking meat alone, which was partly accounted for by the digestive disturbances previously described (1) which resulted in a loss of appetite and diminished intake.

Calcium Balance.—In the preliminary period, the subject K. A. had a slightly negative calcium balance, with a daily intake of 0.369 gm. Sherman has stated that the average calcium requirement for a man is 0.45 gm. per day. It is interesting to note that the mean daily calcium excretion of this subject while receiving the meat diet was 0.44 gm.—the urinary excretion was roughly 3 times the amount excreted in the feces. In the preliminary period, V. S. had a positive balance with an intake of 0.434 gm. daily. His urinary excretion of calcium was considerably less than that of the other subject. Both men had definitely negative balances throughout the periods of meat ingestion. It was also noted that both the calcium excreted in the urine and the total output were greater than the amounts excreted when they were receiving mixed diets. This may have been due to the acid nature of the diet.

The supplementary intake of calcium which was not accurately controlled, had its sources in the drinking water, table salt, and small particles of bone taken with the bone marrow. The drinking water was found to contain approximately 0.01 gm. of calcium per liter. On the assumption that the subjects drank 2 liters of water per day, it would contribute 0.02 gm. of calcium daily or 0.2 gm., for a period of 10 days. Compared with a negative balance of 3.0 gm. of calcium, this would account for only 6 per cent of the loss. Analysis of the table salt used showed a content of 0.004 gm. of calcium in 1.0 gm. of salt. They took between 1.0 and 5.0 gm. of salt per day which would account for between 3 and 6 per cent of the loss. The calcium intake as reported in Table V does not include the calcium taken in water or table salt.

Both subjects received considerable quantities of bone marrow at various times and to obtain it the shafts of long bones were sawed in sections about 6 inches long and cracked open with a hammer. In this process, some splintering occurred and there was a possibility of removing a slight amount of the adjacent bone with the marrow. Marrow supplied in this way was divided in half; one part was eaten, and the other half was sent to the laboratory for analysis. Determinations, made on eleven samples of marrow, showed a variation in calcium content ranging from 0.043 to 0.092 gm. per 100 gm. of marrow. Eight of these determinations showed from 0.050 to 0.076 gm. of calcium per 100 gm. of marrow. As the samples for each period represented fairly accurately the marrow ingested in that period, the errors arising from this source are not significant. Several analyses of marrow that had been carefully removed with a spatula without cracking the bone or scraping the sides showed a mean calcium content of 0.005 gm. per 100 gm. of marrow. The increase in the calcium intake of the subject K. A. in the third and subsequent periods is due mainly to the calcium received in the bone marrow.

In Period 7, V. S. showed a 3-fold increase of calcium in the feces and an apparent increase in the calcium loss. The estimated intake of calcium and phosphorus during this period was undoubtedly much too low, as he ate the meat directly from the bone, sometimes chewing the soft ends of the ribs, while previously he was given meat which had been cut from the bones.

No evidence was obtained which indicated that the loss of calcium from the body was in any way serious. Roentgenograms of the hands, when compared with those of men of approximately the same size who were receiving mixed diets, showed no rarefaction. The loss of calcium, observed during the intensive studies, may not have continued when they ate the meat from the bone.

Phosphorus Balance.—The phosphorus balance in the preliminary periods was positive for both subjects. With the higher intake of phosphorus during the meat periods, it varied from negative to positive. The increased phosphorus ingested was entirely eliminated in the urine. The average daily excretion in the feces decreased during the meat periods. The balance for the entire period of meat ingestion was +2.34 gm. for K. A. and −5.34 gm. for V. S. Throughout the period of observation the blood calcium and phosphorus remained normal (Tolstoi (16)).

SUMMARY AND CONCLUSIONS.

1. The results of the analyses of many samples of meat are presented. They agree in general with the findings in other available reports.

2. Observations were made to determine the efficiency of absorption of the foodstuffs from the intestinal tract. The loss of nitrogen in the feces of K. A. was 4.5 per cent of the intake and for V. S. was 7.5 per cent. The loss of fat for K. A. was 3.0 per cent and for V. S., 9.2 per cent.

3. The use of the meat diets by the two subjects studied did not reduce the efficiency of absorption from the intestinal tract.

4. The nitrogen, calcium, and phosphorus balances of the two men are presented.

5. One subject (K. A.) remained in nitrogen equilibrium while receiving meat with an average daily intake of 19.9 gm. and the other (V. S.) showed a slight negative balance with an intake of 18.7 gm. per day.

6. The daily intake of calcium was from 0.05 to 0.15 gm. The average daily excretion of calcium for one subject was 0.44 gm., and for the other 0.47 gm. Both men showed negative calcium balances when taking meat alone.

7. In spite of the high phosphorus intake, K. A. showed a positive phosphorus balance of only +2.34 gm. for the entire period of meat ingestion, while V. S. showed a balance of −5.34 gm. This

was due to the increased excretion of phosphorus in the urine since the phosphorus in the feces decreased when the meat diet was taken.

8. The influence of the acid nature of the diet on mineral balances has been discussed.

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