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Nitrogen Balances during Recovery from Severe Undernutrition

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The results reported below were obtained on two groups of subjects, Group I in Western Holland in 1945 and Group II in the British Zone of Germany in 1946. Among the objects of the Dutch investigation was the determination of the calorie- and protein-intake levels which would secure optimum nitrogen retention and rapid increase in weight in severely undernourished patients. The later work in Germany had as one of its objects the determination of the calorie and protein-nitrogen intakes necessary to secure nitrogenous equilibrium in subjects who were considerably wasted. They had lived on diets low in calories and protein over a period of about 1 year and were in slight negative nitrogen balance when first observed.

METHODS

The calorie and nitrogen contents of the daily intakes were estimated from food tables; for foodstuffs of British origin McCance & Widdowson's (1942) tables were used. These were supplemented by values given by the Council of British Societies for Relief Abroad (1945) which contained more up-to-date information on wartime foods and especially on those of American origin. A third set of tables prepared by the Netherlands Food Administration (1944) during the German occupation was of great value in determining the composition of foodstuffs of Dutch origin, such as bread baked from locally produced flour, wartime cheeses and certain vegetables not included in the previous tables. Owing to the variations in the proportions of the cereals used in preparing the flour used for bread- and biscuit-making in Holland and for bread-making in Germany, it was necessary to carry out periodical analyses to

obtain values for the nitrogen content of the flour and bread in the diet. In the German part of the work, the nitrogen content of each new diet was estimated by direct analysis to provide a check on the values obtained by tables. It was found that the estimated values for nitrogen content were within $\pm 3\%$ of the values determined by analysis. This close approximation was due to the small number of standard food-stuffs used in the diets and to the constancy of their nitrogen content. No attempt was made to determine experimentally the calorie value of the daily diets.

Urine was collected over thymol and toluene for each period of 24 hr., care being taken to ensure exact timing of the complete sample. After acidification with a few drops of conc. sulphuric acid a portion of each 24 hr. specimen was taken and stored until analysed. Nitrogen was determined by a semi-micro Kjeldahl method using 1 ml. samples of urine and a selenium dioxide-copper sulphate catalyst. Digestion was continued with gentle boiling for at least 2 hr. after the mixture had cleared. No increase in ammonia titre was obtained by longer combustion. Agreement of duplicates was within 1%. Duplicate determinations were not carried out simultaneously. Stools were brought to the laboratory in bed pans and were transferred to large glass jars containing about 250 ml. of water; 100 ml. of conc. sulphuric acid were added slowly with constant stirring. Additional stools were treated in the same way. When the complete sample had been collected, another 250 ml. conc. sulphuric acid were added to the entire mass. After cooling, the material was passed through a fine-wire sieve and distilled water added to give a known volume (approx. 500 ml. for each day's collection). The sample was agitated to ensure thorough mixing. Five portions, each about 10% of the total sample, were taken. These were digested at boiling-point after addition of more sulphuric acid until solution was complete. From each, samples were taken and digested for varying lengths of time with sulphuric acid and catalyst until a constant maximal ammonia titre was obtained. The mean of these values from each portion was taken as the final figure.

Analyses of foodstuffs and diets were carried out by a process similar to that recorded for analysis of faeces.

Details of the composition of the diets are given in the Appendix (p. 217).

RESULTS

In Table 1 the intakes for each balance period are given. The balance periods succeeded each other without any intervening 'free' days. Nitrogen losses in the faeces are set out in Table 2 and the balances for each period are contained in Table 3. Intakes, outputs and balances have been expressed in terms of Cal. and g. N/kg. body-weight. The body-weight used in the calculations was the mean value for each period.

In calculating average body-weights for each balance period, the mean weight was calculated from the daily weighings. In no balance period was the weight change between the first and last day of the period greater than 1.5 kg., in most it was less than 1 kg.

The results for Groups I and II have been expressed graphically in Figs. 1 and 2, where the average daily nitrogen balances have been plotted against the average daily

Table 1. *Dietary data for the two groups of subjects*

Subject	Balance period (days)	Type of diet*	Average total daily intake		Average daily intake/kg. body-weight	
			Calories	Nitrogen (g.)	Calories	Nitrogen (g.)
Group I (Dutch):						
J.	16	H. P.	3150	49.7	76.3	1.20
E.	13	L. P.	2940	25.5	52.6	0.46
	10	H. P.	3180	50.2	55.6	0.88
Z.	11	L. P.	2970	25.4	64.7	0.55
K.	10	L. P.	2980	25.3	59.7	0.51
V.	10	L. P.	2860	23.8	61.1	0.51
F.	10	Mx.	2910	39.6	69.2	0.94
Group II (German):						
Ks.	10	Mx.	1890	10.3	33.8	0.18
	15	Mx.	2100	9.8	37.7	0.18
	10	Mx.	2220	13.3	39.1	0.23
	10	Mx.	2200	14.6	39.1	0.26
	13	Mx.	2520	19.2	43.9	0.33
Lv.	12	Mx.	2190	12.7	39.5	0.23
Wz.	12	Mx.	2000	11.3	37.5	0.21
Kl.	10	Mx.	1700	9.1	31.7	0.17
	15	Mx.	2100	10.0	38.9	0.185
	10	Mx.	2210	13.2	40.6	0.24
	10	Mx.	2190	14.6	40.2	0.27
	13	Mx.	2530	19.2	45.8	0.35
Sc.	10	Mx.	1810	9.0	30.6	0.16
	15	Mx.	2090	9.7	37.2	0.17
	10	Mx.	2210	13.2	39.0	0.23
	10	Mx.	2190	14.5	38.5	0.26
	13	Mx.	2530	19.2	43.9	0.33
Gm.	10	Mx.	1700	8.9	33.5	0.18
	15	Mx.	2080	9.7	41.1	0.19
	10	Mx.	2210	13.2	43.3	0.26
	10	Mx.	2160	14.5	42.6	0.29
	13	Mx.	2520	19.2	49.2	0.37
Gt.	10	Mx.	1700	9.0	34.7	0.18
	15	Mx.	2080	9.6	42.1	0.20
	10	Mx.	2140	13.1	42.5	0.26
Rs.	10	Mx.	1730	9.2	31.1	0.17
	15	Mx.	2090	9.7	38.0	0.18
	10	Mx.	2220	13.3	40.3	0.24
	10	Mx.	2200	14.6	39.9	0.27
	13	Mx.	2530	19.2	45.7	0.35
Gd.	10	Mx.	1720	8.9	35.7	0.20
Bm.	10	Mx.	1710	9.2	34.9	0.19
	15	Mx.	2080	9.7	42.3	0.19
	10	Mx.	2200	13.2	43.9	0.26
	10	Mx.	2180	14.5	43.2	0.29
	13	Mx.	2530	19.2	49.1	0.37
Ru.	10	Mx.	1710	9.1	31.7	0.17

* See Appendix (p. 217).

Table 2. *Average values for faecal nitrogen of the two groups of subjects*

Subject	Faecal nitrogen (g./day)		
	Calculated (10% of intake)	Determined	Value used for calculation of balance
Group I (Dutch):			
J.	5.0	3.3	3.3
E.	2.6	3.4	3.4
	5.0	4.1	4.1
Z.	2.5	1.5	1.5
K.	2.5	—	2.7*
V.	2.4	1.4	1.4
F.	4.0	1.8	1.8
Group II (German):			
Ks.	1.0	2.3	2.3
	1.0	1.2	1.2
	1.3	1.3	1.3
	1.5	1.3	1.3
	1.9	1.4	1.4
Lv.	0.9	2.3	2.3
Wz.	0.9	2.3	2.3
Kl.	0.9	2.3	2.3
	1.0	1.2	1.2
	1.3	1.7	1.7
	1.5	1.7	1.7
	1.9	—	2.0
Sc.	0.9	2.0	2.0
	1.0	1.2	1.2
	1.3	—	1.3
	1.5	—	1.5
	1.9	2.0	2.0
Gm.	0.9	1.2	1.2
	1.0	—	1.2
	1.3	2.0	2.0
	1.5	1.7	1.7
	1.9	2.0	2.0
Gt.	0.9	2.0	2.0
	1.0	1.2	1.2
	1.3	—	1.3
Rs.	0.9	2.0	2.0
	1.0	0.7	0.7
	1.3	—	1.3
	1.5	—	1.5
	1.9	1.9	1.9
Gd.	0.9	—	2.0†
Bm.	0.9	—	2.0†
	1.0	1.5	1.5
	1.3	1.6	1.6
	1.5	1.6	1.6
	1.9	1.6	1.6
Ru.	0.9	1.9	1.9

* This value was obtained from a preceding short balance period with a comparable diet.

† Assumed, as this was the loss obtained in patients Sc., Gt. and Rs. with the same diet as that eaten by Gd. and Bm.

Table 3. *Average nitrogen balances of the two groups of subjects*

Subject	Nitrogen intake (g./day)	Nitrogen output in urine (g./day)	Nitrogen balance		Average body-wt. (kg.)
			g./day	g./kg. body-wt./day	
Group I (Dutch):					
J.	49.7	36.6	+ 9.8	+ 0.237	40.0
E.	25.5	17.6	+ 4.5	+ 0.081	55.9
	50.2	35.9	+ 10.2	+ 0.178	57.2
Z.	25.4	18.0	+ 5.9	+ 0.129	45.9
K.	25.3	17.0	+ 5.6	+ 0.112	50.0
V.	23.8	14.1	+ 8.3	+ 0.178	46.8
F.	39.6	30.0	+ 7.8	+ 0.186	42.0
Group II (German):					
Ks.	10.3	7.8	+ 0.2	+ 0.004	55.9
	9.8	6.9	+ 1.7	+ 0.030	55.9
	13.3	9.7	+ 2.3	+ 0.040	57.0
	14.6	13.0	+ 0.3	+ 0.005	56.2
Lv.	19.2	15.4	+ 2.3	+ 0.041	57.4
	12.7	9.3	+ 1.1	+ 0.019	55.5
	11.3	9.0	0	0	53.5
	Kl.	9.1	6.4	+ 0.4	+ 0.007
10.0		5.8	+ 3.0	+ 0.055	53.9
13.2		8.0	+ 3.5	+ 0.064	54.5
14.6		10.5	+ 2.4	+ 0.044	54.6
19.2		12.5	+ 4.7	+ 0.085	55.2
Sc.	9.0	7.8	- 0.8	- 0.013	56.0
	9.7	7.0	+ 1.5	+ 0.027	56.2
	13.2	9.6	+ 2.3	+ 0.039	56.7
	14.5	11.5	+ 1.5	+ 0.027	57.0
Gm.	19.2	13.8	+ 3.4	+ 0.059	57.6
	8.9	7.2	+ 0.5	+ 0.010	50.6
	9.7	6.6	+ 1.9	+ 0.038	50.5
	13.2	8.7	+ 2.5	+ 0.048	51.0
	14.5	10.6	+ 2.2	+ 0.044	50.8
Gt.	19.2	14.1	+ 3.1	+ 0.061	51.3
	9.0	6.7	+ 0.3	+ 0.007	48.9
	9.6	6.3	+ 2.1	+ 0.042	49.4
Rs.	13.1	8.6	+ 3.1	+ 0.062	50.3
	9.2	7.5	- 0.3	- 0.005	55.5
	9.7	7.3	+ 1.7	+ 0.030	55.0
	13.3	10.0	+ 2.0	+ 0.036	52.2
	14.6	12.4	+ 0.7	+ 0.012	55.0
Gd.	19.2	15.0	+ 2.3	+ 0.040	55.3
	8.9	7.4	- 0.5	- 0.009	45.6
Bm.	9.2	6.8	+ 0.4	+ 0.008	49.1
	9.7	6.4	+ 1.8	+ 0.036	49.2
	13.2	9.2	+ 2.4	+ 0.048	50.1
	14.5	11.1	+ 1.8	+ 0.036	50.4
	19.2	14.4	+ 3.2	+ 0.063	51.4
Ru.	9.1	9.0	- 1.8	- 0.035	54.1

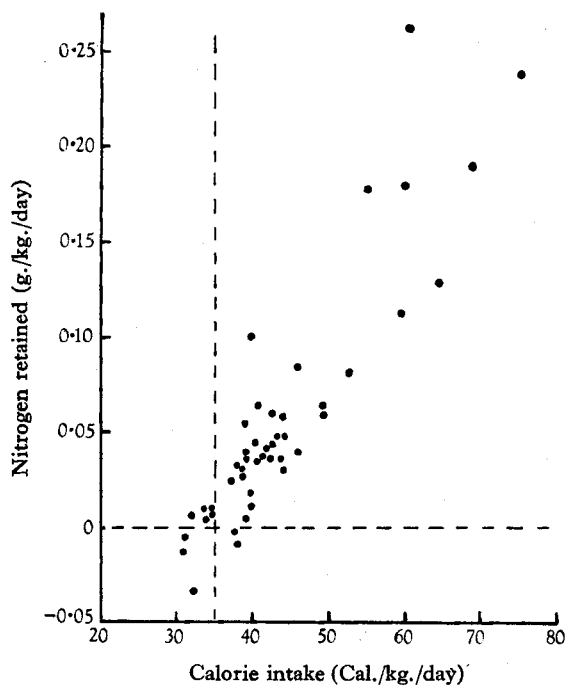


Fig. 1. Relation between calorie intake expressed as Cal./kg. body-weight/day and the nitrogen retention expressed as g./kg. body-weight/day. Balance periods of 10 days and over. For the meaning of the vertical broken line see p. 208.

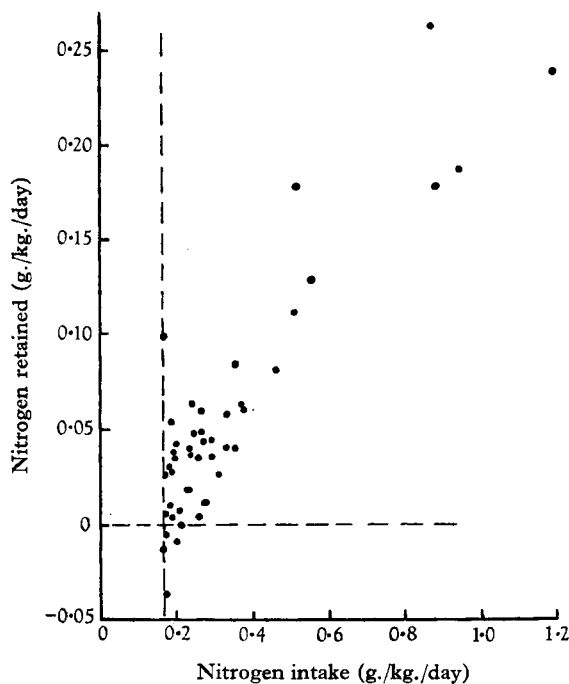


Fig. 2. Relation between nitrogen intake expressed as g./kg. body-weight/day and the nitrogen retention similarly expressed. Balance periods of 10 days and over. For the meaning of the vertical broken line see p. 208.

calorie and nitrogen intakes for each balance period. Fig. 3 has been plotted to show average daily balances against calorie intakes between 30 and 40 Cal./kg. Beside each point is the average nitrogen intake of each period. The relation between calorie intakes above 42.5 Cal./kg. and nitrogen retention has been plotted in Fig. 4. Nitrogen intakes in g./kg./day are indicated beside each point. Table 4 and Fig. 4 contain values obtained during short balance periods of 4-6 days in our Dutch series. These are indicated by \otimes on the graph. Nitrogen intakes are also placed against each point.

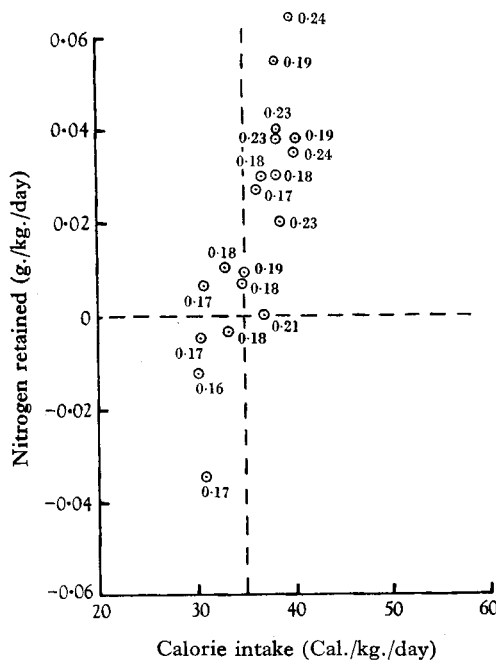


Fig. 3. Nitrogen retention as related to calorie and nitrogen intakes less than 42.5 Cal./kg. body-weight/day and 0.24 g. N/kg. body-weight/day. The sign of the nitrogen balance is determined by the calorie intake. Balance periods of 10 days and over. Figures beside each point indicate the average nitrogen intake in g./kg./day. For the meaning of the vertical broken line see below on this page.

Fig. 1. suggests that nitrogen retention is roughly proportional to calorie intake. The graph, however, indicates that there is a critical calorie intake level around 30-40 Cal./kg. where the level of the nitrogen intake determines the sign and magnitude of the balance. It would appear from Fig. 2 that at nitrogen intake levels above 0.2 g./kg./day the balance is always positive. At high intake levels, those above 0.3 g./kg., the points seem to show a shift to the right indicating that retention at such levels is not proportional to the nitrogen intake.

At the critical lower level of intake of 30-40 Cal./kg./day it would appear (Fig. 3) that it is possible to obtain significant positive nitrogen balances on nitrogen intakes as low as 0.17 g./kg., provided the calorie intake exceeds 35 Cal./kg. Allowing for inherent errors in the balance values it would be justifiable to conclude that nitrogen equilibrium in our emaciated subjects was attained when the calorie intake was no

less than 35 Cal./kg. and the nitrogen intake not less than 0.17 g./kg. If one or other of these intakes were less a negative nitrogen balance might be expected.

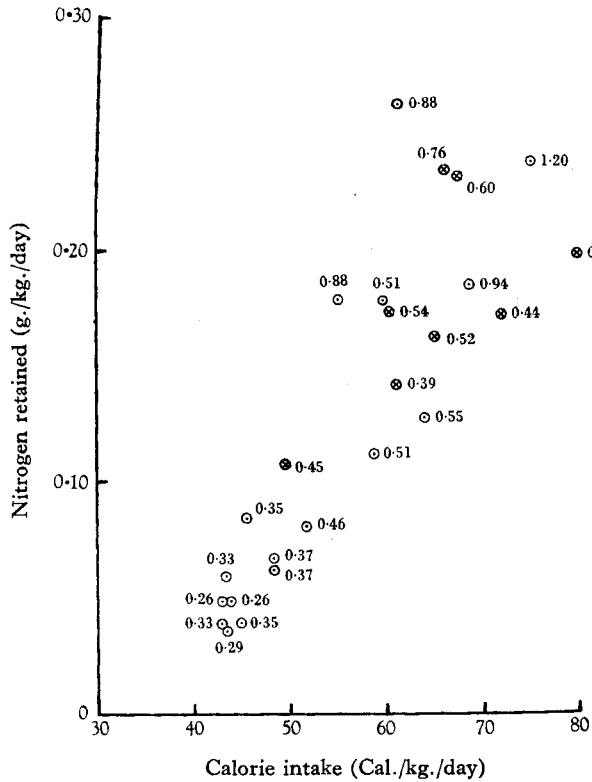


Fig. 4. Nitrogen retention as related to calorie and nitrogen intakes above 42.5 Cal./kg. body-weight/day and 0.24 g. N/kg. body-weight/day. Balance periods of less than 10 days are indicated thus ⊗. The figures beside each point indicate the average nitrogen intake in g./kg./day.

Table 4. Average values obtained in short balance periods (Dutch group)

Subject	Type of diet*	Balance period (days)	Daily intake/kg. body-weight		Nitrogen retained	
			Calories	Nitrogen (g.)	g./kg. body-weight/day	As percentage of nitrogen absorbed
T. Jr.	M.	5	49.6	0.44	0.106	24
T. Sr.	H.	6	62.3	0.38	0.141	37
d. G.	M.	6	66.2	0.58	0.167	29
v. d. H.	H.	6	71.3	0.43	0.156	36
B.	M.	6	62.3	0.55	0.201	37
T.	H.	5	80.3	0.48	0.195	41
v. A.	M.	5	68.1	0.60	0.222	37
Bg.	H. P.	4	67.7	0.76	0.235	23

* See Appendix (p. 217).

Examination of Fig. 4 reveals that at certain retention levels, where there is only a small difference in the calorie intake/kg., there is a wide variation in the nitrogen intake/kg., e.g. at calorie intakes between 67 and 76.5 Cal./kg./day, the nitrogen retention was between 0.23 and 0.24 g. N/kg./day but the nitrogen intakes varied between 0.6 and 1.2 g. N/kg./day. These and other similar examples in the figure suggest that in some diets a considerable proportion of the dietary protein was being used as a source of energy. It is conceivable that the ratio of non-protein to protein calories in the diet may

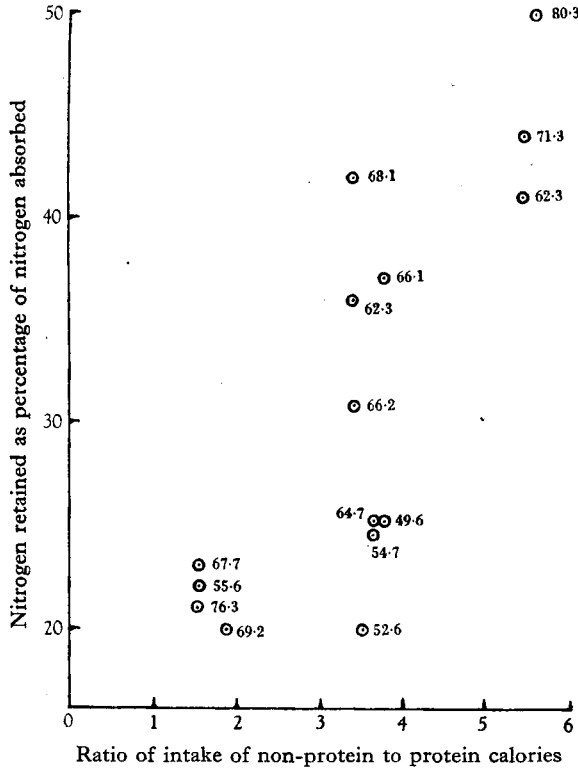


Fig. 5. Relation between intake of protein and non-protein calories and nitrogen retention (Dutch series). The figures beside each point indicate calorie intakes expressed as Cal./kg. body-weight/day.

have had some influence on the level of nitrogen retention. Where there was a high proportion of non-protein calories there may have been a marked protein-sparing action and consequently a better nitrogen retention.

Table 5 gives average nitrogen retentions and intakes of protein and non-protein calories of the two groups of subjects, while Fig. 5 shows the nitrogen retention in Group I (with the eight balance periods in Table 4) on different ratios of non-protein to protein calories in the diet. The calorie intake levels expressed as Cal./kg./day are shown alongside each point. Four periods, where the non-protein/protein ratios were between 1.5 and 2.0, gave retentions between 21 and 23%. These periods where the non-protein/protein ratio was 5.4-5.5 gave retentions between 40 and 50%. The intermediate ratios were around 3.5. In them there was an exceedingly wide range of

Table 5. *Average nitrogen retention and intake of protein and non-protein calories of the two groups of subjects*

Subject	Nitrogen absorbed (g./day)	Nitrogen retained		Energy intake			Ratio of intake of non-protein to protein calories
		(g./day)	As percentage of nitrogen absorbed	Total (Cal./day)	From protein (Cal./day)	From non-protein sources (Cal./day)	
Group I (Dutch):							
J.	46.4	9.8	21	3150	1270	1880	1.5
E.	22.1	4.5	20	2940	650	2290	3.5
	46.1	10.2	22	3180	1290	1890	1.5
Z.	23.9	5.9	25	2970	650	2320	3.6
K.	22.6	5.6	25	2990	650	2340	3.6
V.	22.4	8.3	37	2860	610	2250	3.7
F.	37.8	7.8	21	2910	1010	1900	1.9
T. Jr.	18.9	4.8	25	2360	510	1850	3.7
T. Sr.	16.2	6.7	41	2930	460	2470	5.4
d. G.	27.1	8.3	31	3360	760	2600	3.4
v. d. H.	16.1	7.0	44	2930	460	2470	5.4
B.	26.7	9.5	36	3360	760	2600	3.4
T.	14.4	7.2	50	2930	450	2480	5.5
v. A.	25.5	10.6	42	3090	700	2390	3.4
Bg.	47.4	11.0	23	3170	1280	1890	1.5
Group II (German):							
Ks.	8.0	0.2	3	1890	1260	1630	6.2
	8.6	1.7	20	2100	250	1850	7.4
	12.0	2.3	19	2220	340	1880	5.5
	13.3	0.3	2	2200	370	1830	4.9
	17.8	2.3	13	2520	490	2030	4.1
Lv.	10.4	1.1	11	2190	230	1870	5.7
Wz.	9.0	0	0	2040	290	1750	6.1
Kl.	6.8	0.4	6	1700	230	1470	6.3
	8.8	3.0	3	2100	260	1840	7.2
	11.5	3.5	30	2210	340	1870	5.5
	12.9	2.4	17	2190	370	1820	4.9
	17.2	4.7	27	2530	490	2040	4.1
Sc.	7.0	-0.8	—	1710	230	1480	6.4
	8.5	1.5	18	2090	250	1840	7.3
	11.9	2.3	19	2210	340	1870	5.5
	13.0	1.5	11	2190	370	1820	4.9
	17.2	3.4	20	2530	490	2040	4.1
Gm.	7.7	0.5	6	1700	230	1470	6.4
	8.5	1.9	22	2080	250	1830	7.3
	11.2	2.5	22	2210	340	1870	5.5
	12.8	2.2	17	2160	370	1790	4.8
	17.2	3.1	18	2520	490	2030	4.1
Gt.	7.0	0.3	4	1690	230	1460	6.3
	8.4	2.1	25	2080	250	1830	7.5
	11.8	3.1	26	2140	340	1800	5.4
Rs.	7.2	-0.3	—	1730	240	1490	6.3
	9.0	1.7	19	2090	250	1840	7.4
	12.0	2.0	17	2220	340	1880	5.5
	13.1	0.7	5	2200	370	1830	4.9
	17.3	2.3	13	2530	490	2040	4.1
Gd.	6.9	-0.5	—	1720	230	1490	6.5
Bm.	7.2	0.4	6	1710	240	1470	6.3
	8.2	1.8	22	2080	250	1830	7.3
	11.6	2.4	21	2200	340	1860	5.5
	12.9	1.8	14	2180	370	1810	4.9
	17.6	3.2	18	2530	490	2040	4.1
Ru.	7.2	1.8	25	1710	230	1480	6.4

percentage retentions, 20-42. It was clear, therefore, that while the protein-sparing action appeared to be present in some balance periods with a high non-protein/protein ratio, it would not have accounted for the wide variation in the intermediate-ratio (3.5) diets. The level of calorie intake may have an influence, but obviously other factors must operate.

DISCUSSION

As Beattie & Herbert (1947) have shown that in the early phases of recovery the metabolic level rises with the calorie intake, it seemed possible that a rise in this level might affect adversely the nitrogen retention. Table 6 gives the relevant data. The

Table 6. *Average values for nitrogen retention and basal heat production of the two groups of subjects*

Subject	Basal heat production, Cal./kg. body-weight/day	Nitrogen retention	
		g./kg. body-weight/day	As percentage of nitrogen absorbed
Group I (Dutch):			
J.	31.8	0.237	21
E.	21.6	0.081	20
	30.7	0.178	22
Z.	28.0	0.129	25
K.	25.4	0.112	25
F.	27.5	0.186	21
T. Jr.	26.0	0.106	25
v. A.	30.3	0.222	42
Group II (German):			
Ks.	26.3	0.004	3
	23.7	0.030	20
	22.1	0.040	19
	22.4	0.005	2
	21.4	0.041	13
Kl.	19.7	0.007	6
	23.0	0.055	3
	26.6	0.064	30
	23.9	0.044	17
	22.5	0.085	27
Gm.	22.4	0.038	22
	24.5	0.048	22
	24.8	0.044	17
	22.3	0.061	18
Gt.	21.4	0.007	4
	22.3	0.042	25
	20.9	0.062	26
Rs.	28.2	-0.005	—
	25.0	0.030	19
	24.1	0.036	17
	21.8	0.012	5
	22.8	0.040	13
Bm.	26.5	0.008	6
	23.3	0.036	22
	21.7	0.048	21
	23.6	0.036	14
	21.1	0.063	18

relation of metabolic level to retention for both the Dutch and German groups is given graphically in Fig. 6. It would appear that retention increased with the rise in metabolic level. This relation, however, may be merely a reflexion of the high calorie and nitrogen intakes—absolute retentions rising as more nitrogen is available in the diet. There is, however, no indication that a high metabolic level necessarily leads to a reduction in the absolute values of nitrogen retention. When the metabolic level is plotted against percentage nitrogen retention there is no conclusive evidence that these two factors are causally related (Fig. 7).

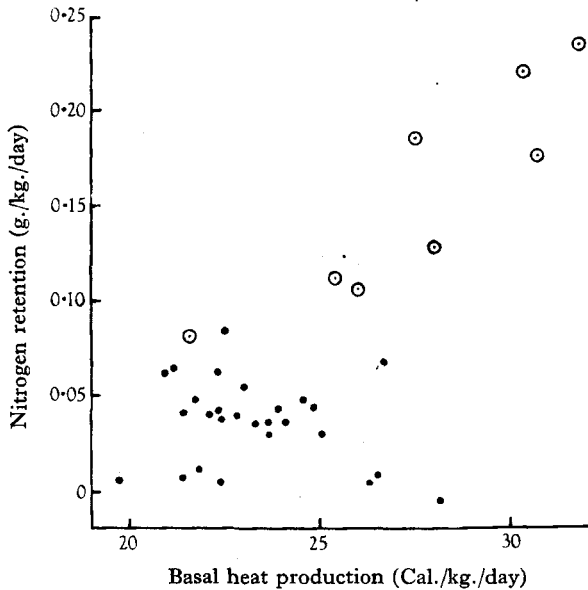


Fig. 6. Relation between the basal heat production expressed as Cal./kg. body-weight/day and nitrogen retention expressed as g. N/kg. body-weight/day. Dutch series ⊙; German series ●. Dutch series includes only first balance period. German series includes all balance periods.

In the German group (Group II) the calorie intakes were confined to a relatively narrow range. It was observed by Beattie & Herbert (1947) that these subjects showed relatively small variations in their metabolic levels. The nitrogen intakes in these men were restricted and consequently errors in estimating their nitrogen retentions were proportionately higher than for Group I where the nitrogen intakes were greater. It is none the less remarkable that the percentage nitrogen retention should show no relation to the non-protein/protein ratio (Fig. 8). Apparently the low calorie- and protein-intake level in these men, coming, as it did, very close to their maintenance levels in the early diet periods, provided a narrow and variable margin for conservation, too narrow for the protein-sparing action of non-protein calories to be apparent.

One further aspect of nitrogen retention seemed worthy of study. It seemed possible that, on the high levels of calorie intake with consequent increase in metabolic level during the early phase of recovery, the carbohydrate utilization might not be complete owing to a relative deficiency in vitamin B₁. Utilization beyond the pyruvate stage might be prevented with a consequent demand for more carbohydrate which

would only be satisfied by the de-amination of amino-acids and the conversion of the carbon skeletons into glycogen. Such an effect would diminish the nitrogen available for retention. Table 7 provides the necessary data. All these patients were on diets in which the main vitamin B₁ intake was derived from skim milk powder of a known vitamin B₁ content and from glucose which had been reinforced by the addition of synthetic vitamin B₁ in the proportion of 1.33 mg./100 g. glucose.

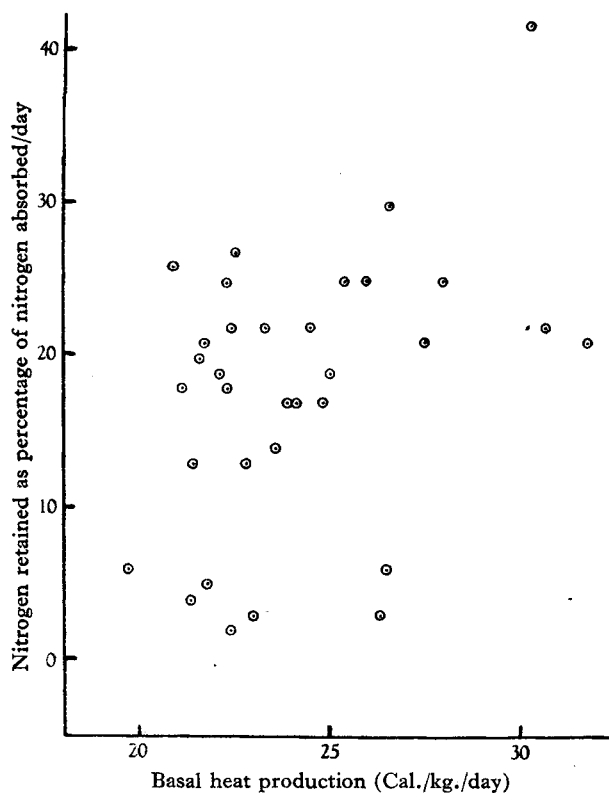


Fig. 7

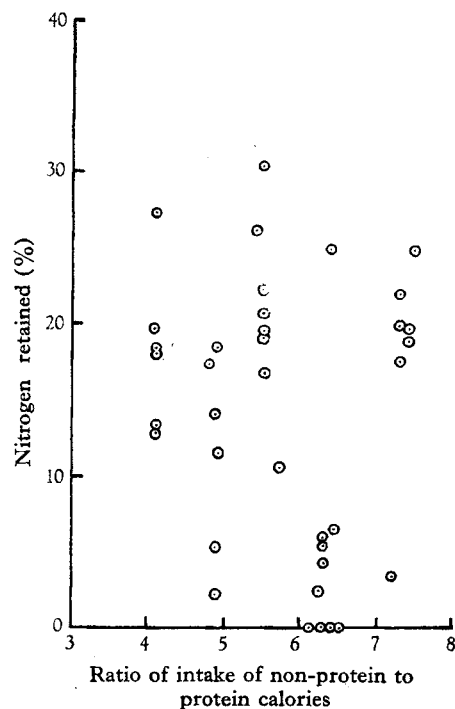


Fig. 8

Fig. 7. Relation between the basal heat production expressed as Cal./kg. body-weight/day and nitrogen retention expressed as percentage of nitrogen absorption (German and Dutch series).

Fig. 8. Relation between intake of protein and non-protein calories and nitrogen retention (German series).

The diets which were relatively poor in vitamin B₁ were those with a high protein content of about 50 g. N/day. The diets richest in vitamin B₁ were those containing only 18 g. N/day. The non-protein calorie intake in the latter group was obtained exclusively from the reinforced glucose. This combination was unfortunate, for the diets with a low non-protein/protein ratio were those with a low vitamin B₁ content and those with a high non-protein/protein ratio were richest in the vitamin. When nitrogen retention as g. N/kg./day is compared with the vitamin B₁ intake expressed either as mg./kg./day or as mg./1000 Cal./day, it would appear that nitrogen retention was not related to the intake of vitamin B₁ (Fig. 9A and B). Examination of Table 7

shows that three of the diets contained less than the Cowgill (1934) standard requirement calculated from the formula

$$\text{Vitamin B}_1 \text{ requirement} = \frac{\text{Calorie value of diet} \times \text{wt. (kg.)} \times 0.0284}{300} \text{ mg.}$$

On these diets the nitrogen retention was high (0.237, 0.178 and 0.235 g. N/kg./day). All other diets contained adequate amounts of vitamin B₁ as judged by this standard.

Table 7. Vitamin B₁ intake and nitrogen retention of the Dutch group of subjects

Subject	Cowgill's (1934) standard requirement for vitamin B ₁ (mg./day)	Vitamin B ₁ intake			Nitrogen retained	
		mg./day	mg./kg. body-weight/day	mg./1000 Cal. intake	g./kg. body-weight/day	As percentage of nitrogen absorbed
J.	1.23	0.35	0.009	0.11	0.237	21
E.	1.57	2.08	0.038	0.68	0.081	20
Z.	1.73	0.35	0.006	0.11	0.178	22
K.	1.30	2.08	0.045	0.68	0.129	25
V.	1.42	2.08	0.042	0.68	0.112	25
T. Jr.	1.30	2.08	0.045	0.68	0.178	37
T. Sr.	1.00	3.56	0.079	1.60	0.106	24
d. G.	1.92	7.44	0.158	2.53	0.141	37
v. d. H.	1.58	5.39	0.106	1.60	0.167	29
B.	1.17	7.44	0.177	2.53	0.156	36
T.	1.84	5.39	0.100	1.60	0.201	37
v. A.	1.01	7.44	0.203	2.53	0.195	41
Bg.	1.32	4.96	0.110	1.60	0.222	37
Bg.	1.88	0.35	0.008	0.11	0.235	23

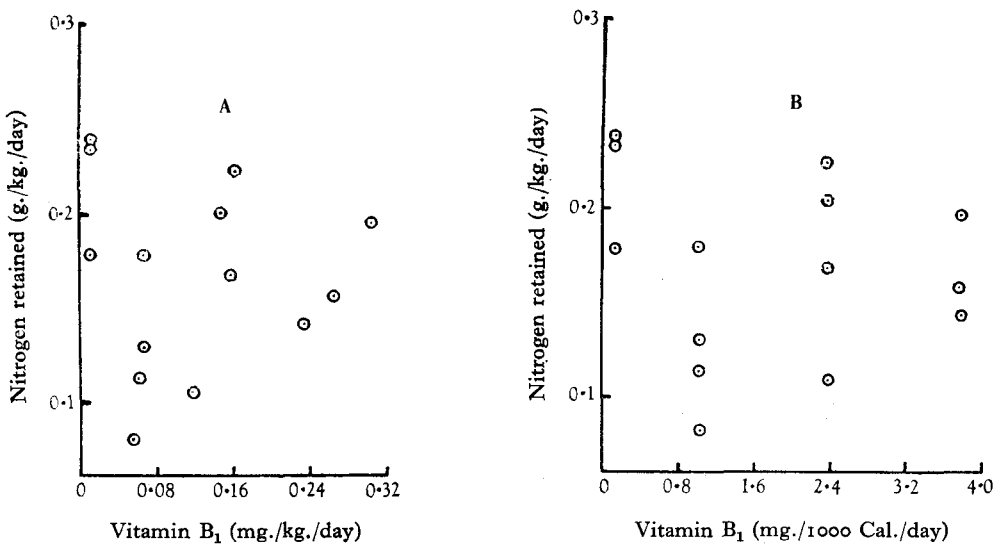


Fig. 9. Relation between vitamin B₁ intake expressed as mg./kg. body-weight/day (A), as mg./1000 Cal./day (B), and nitrogen retention (Dutch series).

The absolute nitrogen retentions of Group I were, with one exception, in excess of 100 mg. N/kg./day, while those of Group II, again with one exception, were below 65 mg. N/kg./day. Although nitrogen retention in absolute values rose with the nitrogen intake, the level of this retention was apparently conditioned by the calorie intake. If the 'efficiency' of the diet is regarded as the percentage of the nitrogen absorbed which is conserved, then those diets which had a high proportion of non-protein calories were more 'efficient'. There is no conclusive evidence that the metabolic level affects the level of nitrogen retention.

SUMMARY

1. In emaciated individuals the sign of the nitrogen balance is positive if the intake is above 35 Cal. and 0.17 g. N/kg. body-weight.
2. The absolute amount of nitrogen retained is apparently related directly to the calorie intake provided the nitrogen intake is above the critical level for nitrogenous equilibrium (0.17 g./kg./day) for the emaciated individual.
3. If the ratio of non-protein to protein calories in the intake is low, nitrogen retention falls. The most efficient ratio for retention at any total calorie intake is apparently above 5 : 1.
4. The metabolic level and the intake of vitamin B₁ have no definite influence on the level of nitrogen retention.

We wish to thank our Dutch assistants for their help in this investigation. Our thanks are also due to Miss A. W. Watts who was responsible for the dietetic work and to the officers of the Public Health Branch of S.H.A.E.F. (Netherlands Mission) and of the Control Commission for Germany for affording us the opportunities to carry out this work. The Medical Research Council provided funds to cover the expenses of the investigation. The Council of the Royal College of Surgeons released two of us (J. B. and P. H. H.) and the University of Cambridge the other (D. J. B.) to make these studies.

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Appendix

*Composition of diets used**High-protein diet (H. P.)*

This diet was designed to provide 50 g. of nitrogen and about 3100 Cal./day with the major part of the nitrogen contained in milk protein.

Constituent	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Skim milk powder	750	43.00	2558
Bread	40	0.47	95
Butter	5	—	39
Eggs	100	2.08	156
Corned beef	100	4.00	244
Potatoes	100	0.22	65
Vegetables	50	0.16	10
		49.93	3167

Low-protein diet (L. P.)

This diet was intended to provide half the nitrogen intake of the high-protein diet (H. P.) with a calorie intake of around 3000 Cal./day.

Constituent	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Skim milk powder	300	17.22	1035
Eggs	100	2.08	156
Corned beef	100	4.0	244
Potatoes	450	0.99	293
Vegetables	100	0.70	53
Bread	40	0.47	95
Butter	5	—	39
Sugar	50	—	197
Glucose	50	—	188
Syrup	50	—	149
Starch porridge	471	—	542
		25.46	2991

Starch porridge had the following composition:

Corn starch	10 g.
Glucose	20 g.
Water to	100 ml.

Milk-powder diets (M.)

Two milk-powder and glucose diets were used. The low-calorie diet provided approx. 1600 Cal. with about 10 g. nitrogen while the high-calorie diet provided approx. 2200 Cal. and 20 g. nitrogen.

	Low-calorie diet			High-calorie diet		
	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Skim milk powder	172	9.85	587	344	19.7	1173
Glucose	197	—	738	281	—	1054
Starch porridge	250	—	288	—	—	—
		9.85	1613		19.7	2227

The glucose used in these and all other diets contained in 100 g. the following added synthetic vitamins:

	mg.
Vitamin B ₁	1·33
Riboflavin	1·33
Nicotinic acid	13·3
Ascorbic acid	33·3

Protein-hydrolysate diet (H.)

This consisted of two foodstuffs—enzymic hydrolysates of meat and casein and glucose.

Constituent	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Protein hydrolysate	150	18·0	458
Glucose	492	—	1845
Starch porridge	545	—	627
		18·0	2930

Mixed diets (Mx.)

The mixed diets given in the Dutch experiments aimed at providing around 3000 Cal. and 25 g. N/day. The animal-protein content of the diet was above 70% of the total protein intake. The following table gives a typical example of such diets.

Constituent	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Milk, standard	1000	5·12	675
Eggs	200	4·16	312
Corned beef	100	4·00	244
Beef	100	3·52	118
Potatoes	150	0·33	97
Vegetables	100	0·20	15
Bread	200	2·34	476
Butter	60	0·05	456
Biscuits	50	0·77	194
Cheese	40	1·86	140
Oatmeal porridge	250	1·68	310
Sugar	15	—	62
Tea	150	—	—
		24·03	3099

The diets used in the German experiments had to be constructed bearing in mind foodstuffs available not only in the institution but in the British Zone for civilian consumption. The diet given below was a standard prison-diet used in the first diet period in the German group.

Constituent	Quantity (g.)	Nitrogen content (g.)	Energy value (Cal.)
Bread	336	3·36	772
Beef	50	1·70	76
Sugar	41	—	162
Flour (mixed rye, oat and wheat)	33	0·51	115
Spring onions	19	0·03	7
Bacon fat	4	—	31
Potatoes (raw and peeled)	444	1·51	386
Lettuce	600	1·02	66
Liver sausage	41	1·18	117
		9·31	1732

Animal protein thus formed about 30% of the total protein intake.

During the second diet period the intake was raised by 500 Cal. from non-protein sources. The protein intake was kept unchanged. In the third diet period the protein intake was raised by adding 25 g. animal protein/day usually as fish (canned pilchards or fresh fish). This raised the animal protein to about 50% of the total protein intake. The calorie intake rose by about 100 Cal. In the fourth diet period the protein content of the diet was raised by 20 g. protein/day usually as fish but the total calorie intake was maintained at the level of the third period. Thus the percentage of animal protein rose to almost 70% of the total protein intake. During the fifth diet period the total calorie intake was raised by about 350 Cal. but the total protein intake was maintained constant at the level of the fourth period.

Nutrition of Domestic Rabbits

1. The Effect of Nutrition on the Carcass Composition of the Rabbit

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It has long been known that different rates of growth when caused by differences in the amount of food eaten profoundly affect the carcass composition of animals. Detailed studies on farm animals have been made by Moulton, Trowbridge & Haig (1922) for cattle, by Vergés (1939) for sheep and by McMeekan (1940*a, b, c*) for pigs. Feeding *ad lib.* favours rapid growth of the late-developing parts of the animal, e.g. anatomically the loin or, chemically the deposition of fat, and therefore gives rapid maturity. Undernutrition stunts these late-developing parts because such food as is available for growth is used preferentially for those parts of the animal which are physiologically most important such as the skull and viscera. Undernutrition therefore produces a 'primitive' or late-maturing type of animal.

Many experiments have been carried out in which the effect of the qualitative nature of the ration on body composition has been investigated by chemical analysis of the whole carcass. In most cases the paired-feeding method was employed. With this technique the faster growing animal of a pair receives less food in relation to its requirements than its mate, and in general tends to lay down less fat in proportion to protein. This is especially true in the later stages of an experiment when one animal of a pair is considerably heavier than the other. Thus Mitchell & Hamilton (1936) found that, with pigs and rats, as the protein level of the ration was raised to about 25%, the proportion of fat to protein laid down decreased. Johnson, Hogan