SUMMARY

Apparatus and technique for the determination of the oxygen consumption and of the carbon-dioxide production of the calf during periods of 30 min. are described. The apparatus is based on that of Tissot (1904), the chief modifications being the considerable increase in its size and the inclusion of a flexible counterpoise for the bell.

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The Nutrition of the Young Ayrshire Calf

3. The Metabolism of the Calf during Starvation and Subsequent Realimentation

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When a calf is affected by acute infantile diarrhoea, 'scouring', there is a marked fall in the amount of nutrients it absorbs from its digestive tract. This is sufficient to cause such negative nitrogen and energy balances that in many severe cases the calf is very close to a state of complete inanition. In farm practice a common and effective method of controlling this type of diarrhoea is to substitute boiled water for the normal milk allowance until the faeces become normal in appearance and then to commence realimentation very slowly. Such a method of control has the effect of substituting complete inanition for the partial inanition that results from diarrhoea. It is of some interest therefore, to study the metabolism of the calf during periods of starvation, and this paper is concerned with experiments designed to study nitrogen, sulphur, energy and mineral metabolism during short periods of starvation.

Much has been published on the effect of starvation on mature animals, especially man, but comparatively little study has been made of the effect of starvation on the metabolism of the really young animal. In the mature bovine, energy metabolism has been studied by Braman (1924), Benedict & Ritzman (1927) and Ritzman & Benedict (1938), and extensive studies of the N metabolism of the fasting steer or cow have been made by Carpenter (1927), Hutchinson & Morris (1936*a*, *b*) and Morris & Ray (1939). The energy metabolism of smaller mature ruminants, more especially sheep, has been studied in detail by Benedict & Ritzman (1931), Brody (1932), Ritzman, Washburn & Benedict (1936), Blaxter (1948) and Marston (1948). Little information, however, is

available on the N metabolism of the smaller ruminants during starvation (Morris & Ray, 1939). The data relating to mature ruminants have been used for comparative purposes in this study.

EXPERIMENTAL

Calves and their treatment

Two Ayrshire bull calves were each subjected to two periods of starvation. The original plan was to study starvation and recovery in each animal when at a normal weight for its age and also following a period of undernutrition when its weight would be only 50 % of that which could be expected for its age. The object of such a design was twofold. Firstly, scouring in calves tends to be chronic before an acute stage is reached, and thus undernutrition for a period of time before almost complete inanition is a common phenomenon. A study of inanition following undernutrition, therefore, seemed desirable. Secondly, it has been shown by Marston (1948) that the minimal level of metabolism is reached more quickly when animals are starved following low levels of food intake than when starved following long periods of overnutrition or optimal nutrition. This may be related to the concept of a minimal base level of heat production (Forbes & Kriss, 1932; Forbes & Swift, 1941) and obviously is of considerable importance. Unfortunately one calf (no. 5) became ill during the course of the pre-starvation period for the low plane of nutrition experiment and this experiment was abandoned. The animal on slaughter was found to have greatly enlarged lymphatic glands. Metabolism results on this animal during infection are not included in this paper.

Details of the calves and of their experimental treatment are given in Table 1. The animals were confined in metabolism crates (Blaxter & Wood, 1951), the routine of feeding and collection of urine and faeces being essentially as described in that paper.

Table 1. *Details of the calves and of their experimental treatment*

Calf no.	Body-weight (kg.)	Age at commencement (days)	Experimental period (days)					
			Normal calorie intake			Low calorie intake		
			Preliminary	Starvation	Recovery	Preliminary	Starvation	Recovery
4	33·5	6	12	4	10	12	4	12
5	34·6	6	18	4	12	14	2	2

The calves were weighed every 2nd morning throughout the experiment except during the starvation periods, when they were weighed every morning.

The pulse rates of the calves were determined twice daily throughout the experiment. Additional records were taken during the starvation periods, coinciding with the respiratory exchange determinations.

Diet

The liquid diet, which was given throughout, was very similar to that described by Blaxter & Wood (1951). The main difference was an increase in its fat content from 39·2 to 42·0 g./l. and a reduction in the glucose content from 14·8 to 14·0 g./l. The amounts given to each calf are shown in Table 2.

Table 2. *Daily quantity of milk diet given to the calves during preliminary and recovery periods*

Calf no.	Normal calorie intake (kg.)	Low calorie intake (kg.)
4	3·8	3·0
5	4·0	3·2

During starvation periods, water was given instead of milk in quantities sufficient to maintain the urine volume at the pre-starvation level.

Analytical procedure

Diet. The digestibility of the dry matter, fat and total N of the diet was determined before and after starvation.

Urine. Analyses of urine were made at 2-day intervals, and daily throughout the period of fasting. Urinary sulphur was determined throughout the experiment. The urinary excretions of calcium, magnesium, sodium, potassium, chlorine and phosphorus were determined on 2-day samples of urine collected during the three periods of each experiment.

The following analytical methods were used:

Total N (Kjeldahl); urea (Van Slyke & Cullen, 1910); ammonia (Van Slyke & Cullen, 1910); creatine and creatinine (Folin, 1914); uric acid (King, 1946); allantoin (Young & Conway, 1942); purine bases (Krüger & Schmid, 1905); heat-coagulable protein (Kjeldahl); total acetone (Van Slyke, 1920); chloride (Volhard, 1878); potassium (Eden, 1943); sodium (Butler & Tuthill, 1931); calcium and magnesium (McCrudden, 1911–12); total sulphur (S. R. Benedict, 1909); inorganic sulphate, ethereal sulphate and neutral sulphur (Folin, 1905–6).

Faeces. Total N (Kjeldahl); total fat, neutral fat plus unsaponifiable residue and soaps (Saxon, 1914) were measured.

Respiratory exchange

In each of the periods of fasting, the respiratory exchange was determined on thirteen occasions, each determination being made for a 30 min. period using apparatus and technique previously described (Blaxter & Howells, 1951). On each occasion the environmental temperature was maintained as close to 20° as possible, the range being 18–21°.

RESULTS

General behaviour of the animals during fasting and when given subnormal quantities of milk

During the preliminary period, when the two calves were given quantities of diet commensurate with normal gains in body-weight, both were very lively and tended to be highly excitable, especially at feeding times. They would stand and play with their harness or their tails for quite long periods. During the first 2 days of starvation this

behaviour pattern continued, the excitability at feeding time still persisting although they were given nothing but water. Later they became more lethargic, but could hardly be called weak. When they were again placed on the normal level of feeding they recovered within a few days. During the period when the subnormal quantity of milk was given, however, activity declined markedly and the animals developed a craving for roughage, a dietary component excluded from their diets. When calf no. 4 was starved on the second occasion, chewing of the walls of the metabolism crate and of his harness increased, and on one occasion he swallowed the whole of the rubber straps supporting his faeces bag. This caused no distress, and in the subsequent recovery period he was noted to have been ruminating using the fragments of rubber tubing which he had previously ingested. At slaughter, 300 g. of chewed rubber were recovered from the rumen of this calf, and throughout the final period rubber appeared in the faeces, invalidating the determinations of carbohydrate by difference methods. Calf no. 5 also consumed small amounts of rubber tubing during the period of subnormal food intake but, although small quantities appeared in the faeces, the rumen at slaughter contained only fragments, obviously from the ends of the straps.

The behaviour of the calves on realimentation is of some interest. Realimentation of calf no. 4 was slow; he was given only half his normal quantity of diet on the 1st day following starvation, and it was noted that his urinary N remained elevated. It was thought that this might be due to the subnormal feeding, so calf no. 5 was given the whole quantity of diet immediately after the starvation period. The result was profuse diarrhoea, which lasted for several days. The same type of reaction in a mild form was shown by calf no. 4 during the second period of realimentation, when two-thirds of the normal allowance of diet was given on the day following starvation. Such alimentary disturbances after fasting are common in man (Lusk, 1928).

Body-weight

Table 3 summarizes the data for both calves. The weights of the calves on commencement of the second period of starvation were 37·95 and 41·25 kg. If the rates of gain in the preliminary period are regarded as normal, these animals should have

Table 3. *Mean daily gains or losses of weight by the calves, calculated from regression analysis of individual weights (g./day)*

	Normal calorie intake			Low calorie intake		
Calf no.	Preliminary period*	Starvation period	Recovery period*	Preliminary period*	Starvation period	Recovery period*
4	307 ± 47	−685	307 ± 42	105 ± 52	−525	130 ± 82
5	302 ± 14	−525	306 ± 26	160 ± 64	−600†	—

* Value with its standard error.
† 2 days only.

reached such weights at 27 and 31 days respectively. Their actual ages were 45 and 56 days. Thus calf no. 4 was retarded in growth by 18 days in 45, or 40 %, and calf

no. 5 by 25 days in 56, or 45 %. These retardations in growth are comparable to those observed in scouring calves. The gain in weight of the animals was not altered significantly by the interpolation of a period of starvation between two periods in which the level of feeding remained constant. There appears, therefore, to be relatively little adaptation to starvation in young animals as judged by economy in body gain following it.

In each instance, the loss of weight in starvation was severe, and there appeared to be no difference which might be judged significant between the weight losses in the two periods. The mean daily loss in weight during three experiments (second period for calf no. 5 excluded) was thus 578 ± 53 g./day.

Pulse rate

Table 4 summarizes the data. Firstly, it will be seen that the lower plane of nutrition was associated with lower pulse rates, even with calf no. 5 during the time in which he was ill. Secondly, starvation when the animal was in normal health resulted in a marked fall in pulse rate to levels in the region of 60 beats/min. The fall was naturally

Table 4. *Mean pulse rates of the calves (beats/min.)*

Calf no.	Calorie intake	8 days before fasting	First 2 days of fast	Second 2 days of fast	2–10 days following fast
4	Normal	94·6	77·5	59·7	86·7
5	Normal	89·8	73·0	64·7	88·3
4	Low	77·8	66·0	59·0	75·1
5	Low	80·9*	81·2*	—	—

* Calf abnormal.

greater when the animal had previously been maintained on a diet containing an adequate supply of energy. Lastly, almost complete recovery of the pulse rate occurred on realimentation. The detailed results plotted in Fig. 1 show the mean fall of pulse rate and return to normality for three complete periods of starvation. In this and subsequent graphs the results for calf no. 5 in the second starvation experiment have been specifically excluded. It may be concluded that during starvation pulse rate drops very markedly, reflecting the fall in the animal's metabolism. The relation between heat production and pulse rate is discussed later.

Digestibility of the diet and the excretion of faeces during starvation

The results are shown in Table 5. There was no significant change in the apparent digestibility of the diet following the first experimental starvation period. With calf no. 4 on the normal level of feeding, the digestibility of the diet increased, but the remaining results showed a decline in digestibility associated with slight diarrhoea on realimentation. These digestibility coefficients are comparable with those that may be calculated from the data of Tomme & Taranenko (1939), where the mean 'digestibility' of dietary energy was 95·1 %, but they are much lower than the earlier German results summarized by Schneider (1947), which indicate 98 % digestibility of the organic

matter and complete digestibility of the ether extractives. It is of some interest, however, that the apparent digestibility of the nitrogen of dried skim milk is of the same order as the true digestibility of the nitrogen of dried skim milk in the rat (Henry, Kon, Lea & White, 1948). Faeces continued to be excreted during the fasting period, and the amounts collected were surprisingly large. The mean daily excretion of dry matter is shown in Table 6.

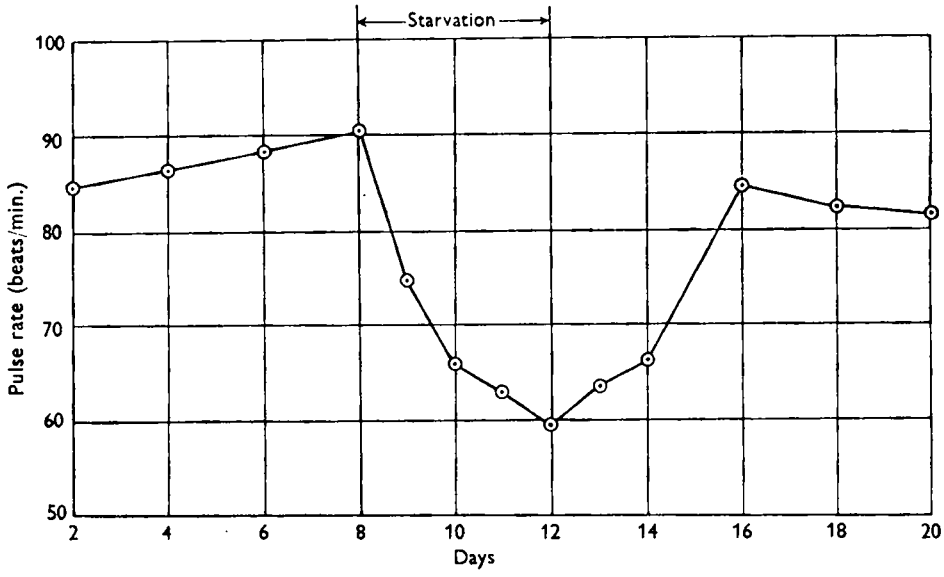

Fig. 1. Effect of starvation on pulse rate in calves.

Table 5. *Mean coefficients of apparent digestibility of the diets before and after starvation*

Calf no.	Level of feeding	Period	Apparent digestibility: Total dry matter (%)	Total fat (%)	Total N (%)
4	Normal	Preliminary	95·7	98·8	89·9
		Recovery	98·3	97·5	98·2
4	Low	Preliminary	96·7	97·1	95·1
		Recovery	94·8	93·5	93·2
5	Normal	Preliminary	95·3	93·2	90·4
		Recovery	92·3	88·7	87·4
Mean of three experiments		Preliminary	95·9	95·4	91·8
		Recovery	95·1	93·2	93·0

Table 6. *Mean daily excretion of dry matter in the faeces during feeding and during starvation (g.)*

Period	Calf no. 4: Normal ration	Calf no. 4: Low ration	Calf no. 5, normal ration	Mean
Preliminary	22·1	13·3	25·2	20·2
Starvation	8·1	13·7	10·1	10·7
Recovery	8·6	20·9	41·9	23·8

The faeces during starvation looked like normal faeces but tended to be more firm. The mean composition of the faeces during feeding and fasting is shown in Table 7.

The higher content of dry matter in starvation faeces is shown in Table 7 and it will also be noted that the composition of the dry matter of the faeces was comparable in both periods. This suggests that the starvation faeces were mainly undigested food

Table 7. *Mean dry-matter content, and mean composition expressed as percentage in dry-matter, of the faeces of the calves during feeding and during starvation*

	Normal	Starvation
Dry matter (%)	18·8	30·7
Total lipids	39·6	39·6
Soaps	25·0	23·1
Neutral fat, free fatty acids and unsaponifiable residue	14·6	16·5
Ash	20·4	19·9
Total N	5·4	5·1

residues and not metabolic products secreted into the gut during starvation. Some fraction of both the normal and starvation faeces must consist of metabolic products, but its magnitude cannot be assessed from the present results.

It may be concluded therefore that a period of starvation does not affect the digestibility of the dry matter, fat or total N of the diet subsequently determined, provided of course that the animal is not severely affected by diarrhoea.

Nitrogen balance and the biological value of the dietary nitrogen

Table 8 summarizes the N-balance results. The total urinary N of calf no. 4 increased during both periods of starvation. With calf no. 5 there was no spectacular change in N excretion in the urine on fasting. In this calf, however, the first few days of realimentation were associated with slight diarrhoea, and urinary N rose markedly. The total loss of N in the urine during starvation was marked. Table 9 besides summarizing

Table 8. *Nitrogen-balance results for the calves during feeding and during starvation (g./day)*

Calf no.	Level of feeding	Period	Length of period (days)	Intake	Excretion		Balance
					Faeces	Urine	
4	Normal	Preliminary	12	16·53	1·67	6·46	+8·40
		Starvation	4	Nil	0·49	9·41	−9·90
		Recovery	4	17·20	0·43	9·47	+7·30
		Recovery	6	17·76	0·25	7·85	+9·66
4	Low	Preliminary	12	13·35	0·65	7·39	+5·31
		Starvation	4	Nil	0·60	10·19	−10·79
		Recovery	4	12·42	0·60	8·88	+2·94
		Recovery	8	13·81	0·94	8·31	+4·56
5	Normal	Preliminary	14	17·27	1·65	7·33	+8·29
		Starvation	4	Nil	0·49	7·11	−7·60
		Recovery	4	15·57	2·62	10·36	+2·59
		Recovery	8	17·91	2·25	7·83	+7·83

the results on these calves, includes for comparison results obtained on other animals by different workers.

During fasting the N katabolism of the young calf is more than twice as intense as that of the cow at maturity and much greater than that of the sheep and of the goat, small ruminants of similar size. This is in substantial agreement with the endogenous N metabolism and basal energy metabolism results previously published (Blaxter & Wood, 1951) and emphasizes the intensity of metabolism in the calf.

Table 9. *The loss of body nitrogen during starvation in the young calf compared with that observed in other animals*

Animal	Loss of N (mg./kg. body-weight/day)	Reference
Young calf	259	Present work
Sheep	152	Morris & Ray (1939)
Goat	162	Morris & Ray (1939)
Pig	60	Voit (1901)
Cow	90	Hutchinson & Morris (1936*a*), Morris & Ray (1939)
Steer	69	Carpenter (1927)

The first 4 days of realimentation, which are shown separately in Table 8, indicate that urinary N remained high, and N retention low, in this period. Part of this in two instances was probably due to the occurrence of the slight alimentary disturbances. In the remaining instance, calf no. 4 in the first period of starvation, no diarrhoea occurred and thus such an explanation is untenable. The increase in urinary N excretion on this occasion was all at the expense of urea N and was associated with a rise in urinary ketone excretion (see below). It would appear therefore that, following a fast, excessive deamination of amino-acids for meeting energy demands takes precedence over replacement of lost tissue N. Alternatively, fasting may result in preferential demands in the subsequent period of realimentation for one particular amino-acid.

The utilization of the dietary protein was studied by calculation of the biological values of the ingested total N using the Thomas-Mitchell procedure (Thomas, 1909; Mitchell, 1923–4). Endogenous N was estimated by using the factor of 80 mg. endogenous N/kg. body-weight, and metabolic faecal N was calculated using the factor of 2·0 g. metabolic faecal N/100 g. dry matter excreted (Blaxter & Wood, 1951). The mean biological value before starvation was 72·9 and in the second period following starvation, excluding the first 4 days, it was 68·3. The difference, however, was not statistically significant (difference $= 4{\cdot}6 \pm 1{\cdot}57$). The low level of food ingestion appeared to reduce the biological value of the protein.

The biological value of the proteins of dried skim milk in these experiments was thus about 70. In growing rats, the biological value of the proteins of dried skim milk is about 84 (Fairbanks & Mitchell, 1935; Sumner, 1938; Swaminathan, 1937*a*, *b*; Henry, Houston, Kon & Osborne, 1939; Henry *et al.* 1948). In adult man, a value of 74 has been obtained (Bricker, Mitchell & Kinsman, 1945), and in mature rats, a value of 78

(Sumner, 1938). In all these experiments the level of protein in the diet was 8 % or, with mature animals, as low as 5 %. In the present work, however, the calves were given a diet which contained 19·6 % protein on a dry basis, and 14·9 % of the total calories came from protein. When two calves were given a diet containing 25·7 % protein in the dry matter, or 19·8 % of the total calories as protein, biological values of 47·9 and 44·8 were obtained. On a diet containing 20·6 % protein in the dry matter, or 16·9 % of total calories as protein, the mean biological value was 59·7 (Blaxter & Wood, unpublished observations). These results are very comparable to those obtained by Hamilton (1938) on rats given diets containing different percentages of protein when the source of protein was dried whole egg.

Maximal biological values were thus not obtained owing to the level of protein intake having been too high, and consequently, deamination rather than storage of protein occurred. Cow's milk has a protein content of 27–30 % of the dry matter or, alternatively, protein calories constitute 23–26 % of the total calories. It would be expected that the percentage retention of N absorbed by the calf receiving its dam's milk would be much lower than could be attained on a milk with a low protein content. In this respect the conclusion of Blackwood, Morris & Wright (1936) that, relative to its N content, cow's milk is markedly deficient in calcium and phosphorus for the young calf may perhaps be modified to read that cow's milk contains an excess of N relative to Ca and P for the nutrition of the young calf, or, alternatively, cow's milk as a sole diet for the calf is grossly deficient in energy if protein retention is taken as a measure of dietary adequacy.

Distribution of nitrogen in the urine

The analytical results obtained on each calf are summarized in Table 10. Fig. 2 shows the mean changes in distribution in more detail and is referred to later.

Table 10 and the average results in Fig. 2 show that urea excretion increased during starvation. This was marked with calf no. 4 on both occasions but was negligible with calf no. 5, a result in agreement with the N-balance results previously discussed. Fig. 2

Table 10. *Mean daily excretion of nitrogen in different nitrogenous metabolites in the urine of the calves during feeding and during starvation* (*g.*)

	Calf no. 4 Level of feeding						Calf no. 5 Normal level of feeding		
	Normal			Low					
Metabolite	Preliminary period (8 days)	Starvation period (4 days)	Recovery period (10 days)	Preliminary period (6 days)	Starvation period (4 days)	Recovery period (10 days)	Preliminary period (10 days)	Starvation period (4 days)	Recovery period (8 days)
Urea	4·62	7·34	6·11	4·68	7·10	4·74	4·80	5·00	3·69
Ammonia	0·39	0·90	0·89	1·01	0·58	1·18	0·66	0·55	1·27
Total urea and ammonia	5·01	8·25	7·00	5·69	7·69	5·92	5·46	5·55	4·96
Creatinine	0·365	0·379	0·285	0·232	0·236	0·229	0·467	0·379	0·441
Creatine	0·124	0·214	0·098	0·125	0·197	0·232	0·108	0·377	0·252
Uric acid	0·038	0·057	0·031	0·026	0·048	0·029	0·037	0·048	0·042
Purine base	0·131	0·148	0·193	0·216	0·206	0·252	0·146	0·138	0·211
Allantoin	0·606	0·520	0·373	0·476	0·569	0·583	0·605	0·507	0·645
Total purine	0·775	0·725	0·597	0·718	0·823	0·864	0·788	0·693	0·898
Residual N	0·729	0·117	0·233	1·039	1·227	2·101	0·483	0·186	1·857

shows that urea excretion fell slowly following the fast, again in agreement with the results for total N metabolism. Ammonia excretion was not markedly affected by starvation. This differs from the effect of starvation in man (Cathcart, 1907) where an increase in ammonia excretion counteracts the marked acidosis.

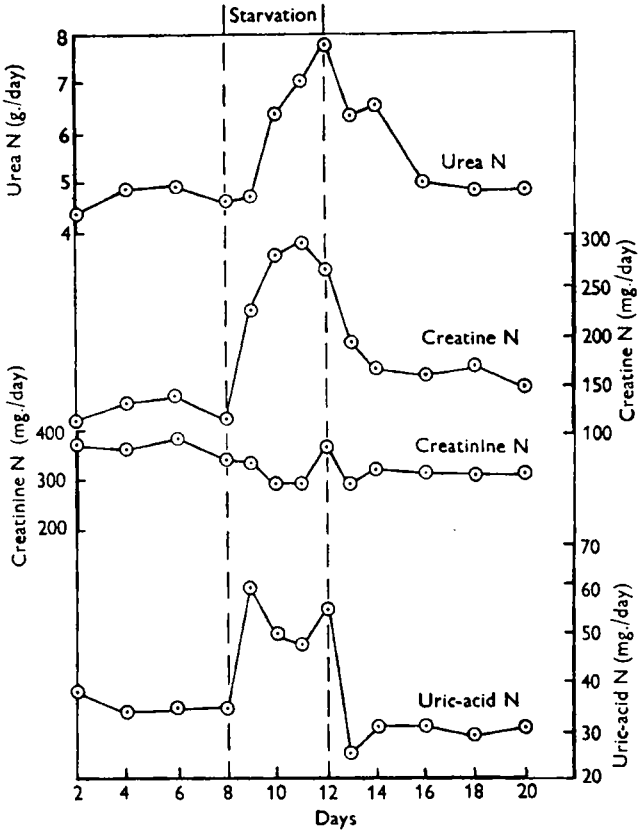

Fig. 2. Effect of starvation on the urinary excretion by calves of urea, creatine, creatinine and uric acid.

The excretion of creatinine declined very slightly throughout, as shown in Fig. 2, there being a slight fall in the recovery period of calf no. 4 in the first starvation experiment, a change of doubtful significance. This is in agreement with Folin's (1905) contention of a constant endogenous metabolism and constancy of creatinine elimination. Creatine excretion rose when the calves were starved. This was true even of calf no. 5, which showed no pronounced change in total N excretion during fasting. The purine bases showed no constant change during starvation, allantoin N did not change, and uric-acid excretion increased, during the starvation period in both animals although the amount of N involved was small. The total purine-N excretion was not affected by starvation, the small variation being commensurate with the day-to-day variation in urinary metabolite excretion met in such studies. The uric-acid N excretion is also shown in Fig. 2.

The residual N, which represents largely amino-N, with the small amounts of N present as protein (albumin) in amines and other compounds, was variable. This was undoubtedly partly due to analytical errors, since the determination of this fraction involves eight separate determinations of N or N-containing compounds. Nevertheless, it was clear in the second experiment with calf no. 4, and more especially in the recovery period of this experiment, as well as in the recovery period of the first experiment with calf no. 5, that the residual N was unduly large. Part of this was traced to an excretion of a heat-coagulable protein. Analytical data were not obtained throughout, but analyses of the urine of calf no. 5 during the preliminary period on the low-calorie diet indicated that up to 1·2 g. N, equivalent to 7·5 g. protein, were being lost daily by the kidneys in the form of a heat-coagulable protein. In any case, there was no indication that any major fraction of the urinary N of starvation was present in the undetermined moiety.

Table 11 compares the N distribution during starvation of the urine of the calf with that of the adult ruminant.

Table 11. *Distribution of nitrogen in different nitrogenous metabolites in the urine of starving calves, goats, sheep and cows expressed as percentages of total urinary nitrogen*

Metabolite	Calf	Goat*	Sheep*	Cow*
Urea	72·49	64·42	57·56	30·39
Ammonia	7·57	3·64	3·87	31·77
Total urea and ammonia	80·06	68·06	61·43	62·16
Creatinine	3·70	6·12	7·88	7·35
Creatine	3·06	3·63	4·86	4·77
Uric acid	0·57	1·08	1·44	0·58
Purine base	1·83	6·16	4·66	2·14
Allantoin	5·95	11·12	18·41	14·24
Total purine	8·35	18·48	24·51	16·96

* Results of Morris & Ray (1939) recalculated and expressed in a form suitable for comparing with present results for calves.

The table shows that the starvation metabolism of the calf differs from that of the cow mainly in that the percentage of creatinine N and total purine N of the total N is only half that found in the same species at maturity or in mature ruminants of similar body-weight. The urea and ammonia N constitutes a considerably greater percentage of the total N in the calf than in mature ruminants. Before discussing this further it is of interest to compare the endogenous N excretion and the N excretion during starvation of the calf, as shown in Table 12. The endogenous N metabolism results in Table 12 are those of Blaxter & Wood (1951); the protein metabolism results are those determined in the preliminary periods of the three present experiments. It is clear that the starvation metabolism must have included the endogenous N metabolism and that the difference between starvation metabolism and endogenous metabolism represents the breakdown of body tissues to provide energy for vital processes during energy deprivation. Of the total increase in N excretion above the endogenous level, 91·1 % was thus due to the excretion of urea and ammonia and 2·8 % to the excretion of creatine. It is probable that these values may be too low, since it was found that total energy metabolism declined during fasting (see below) and thus the endogenous metabolism of the

Table 12. *Nitrogen metabolism of the calf during starvation, its endogenous metabolism and urinary nitrogen excretion during protein ingestion*

Metabolite	Endogenous N excretion* (mg./kg. body-weight/day)	N excretion during starvation (mg./kg. body-weight/day)	Urinary N excretion during protein feeding (mg./kg. body-weight/day)
Total N	81·9	245·6	203·0
Urea	33·2	178·1	129·3
Ammonia	14·5	18·6	19·1
Total urea and ammonia	47·7	196·8	148·4
Creatinine	10·1	9·1	9·8
Creatine	2·9	7·5	3·2
Uric acid	1·3	1·4	0·9

* Results of Blaxter & Wood (1951) recalculated.

calves may also have declined. The slightly lower creatinine excretion per kg. body-weight during fasting adds weight to this suggestion.

This large increase in N excretion during fasting appears compatible with an hypothesis that tissues comparable in composition to muscle substance are katabolized. Table 13 compares the composition of muscle with that of the excess N in the urine, and it will be noted that the katabolism of 0·48 g. muscle/kg. body-weight would yield the same quantity of total N and creatine N and slightly less N as urea and ammonia than was found in starvation.

Table 13. *The composition of muscle (Lusk, 1928) and the increase in the nitrogen excretion during starvation over the endogenous level in the calf*

Constituent	Amount present in muscle (%)	Amount in 0·48 g. muscle* (mg.)	Excess urinary N (mg./kg. body-weight)
Protein N	3·2	153·9	149·1†
Creatine N	0·096	4·6	4·6
Total N	3·4	163·7	163·7

* See text above.

† Sum of urea and ammonia N (amino-N not estimated).

It may be noted from Table 12 that the katabolism of exogenous protein within the body was associated with a small elevation of creatine elimination (1 %) but mainly with an increase in urea and ammonia elimination. It is possible that this small increase in creatine excretion may be due to the dried skim milk in the diet having contained creatine N amounting to 1 % of the total N (Bleyer, 1930).

The differences between the N distributions in the urine of the animals as shown in Table 11, would appear therefore to be entirely due to a proportionately greater katabolism of body protein in the young calf, thus 'diluting' the endogenous moieties of the total N excretion.

The diet used for the experimental calves was virtually purine-free, and yet starvation

did not increase purine excretion, save for a small increase in uric-acid elimination. The same conclusion may be drawn from the calculation that the increased excretion of N above the endogenous level during starvation can be accounted for to the extent of 94 %, leaving only 6 % to be accounted for in terms of purine and residual N. This suggests that in the calf during fasting there is no extensive katabolism of nucleoprotein materials and that the cells of the body remain intact. Morris & Ray (1939) and Hutchinson & Morris (1939*b*) have concluded that in the ruminant there is a marked reduction of nuclear cell metabolism during fasting. These conclusions were based on the reduction of purine-N excretion in the urine when ruminants were starved after having received diets containing normal foods and hay. Such diets are not purine-free and the reduction Morris and his colleagues noted merely reflects a decrease in the exogenous excretion of nucleic-acid derivatives. It is of some interest to compare the distributions of purine N in the urine of the calves and of mature cows. This comparison is shown in Table 14. It will be noted that the calf excretes a slightly higher percentage of its total purine as purine bases than the mature animal. It is also evident, as previously emphasized, that while the mean daily excretion of purine N remains nearly constant when the calf is starved the proportion present as uric acid increases.

Table 14. *Mean percentage distribution of the purine nitrogen in the urine of the calf, cow and goat before and during starvation*

	Calf		Cow*		Goat*	
Constituent	Before starvation	During starvation	Before starvation	During starvation	Before starvation	During starvation
Purine base	22	22	12	15	14	29
Uric acid	4	7	5	3	6	6
Allantoin	74	71	83	82	80	65
Mean daily excretion of purine N (mg.)	760	747	—	—	—	—

* Morris & Ray (1939).

Excretion of sulphur in the urine and the partition of the urinary sulphur

The mean results are shown in Table 15. It is clear from this table that an increase in total S excretion occurred during starvation, smaller in calf no. 5 than in calf no. 4, a result in agreement with the N-metabolism results. The partition of the urinary S

Table 15. *Total urinary excretion of sulphur by the calves and the nitrogen:sulphur ratios during feeding and during starvation*

	Calf no. 4 Level of feeding		Calf no. 5 Normal level	
Period	Normal	Low	of feeding	Mean
	Total excretion of S (mg./day)			
Preliminary	323	413	326	370
Starvation	611	607	403	541
Recovery	476	394	229	366
	N:S ratio			
Preliminary	22·7	19·0	22·0	—
Starvation	16·6	16·7	17·7	—

recorded in Table 16 shows that the major part of the increase in excretion occurred in the inorganic fraction, and that the neutral fraction and the ethereal-sulphate fraction remained constant.

Table 16. *Mean values for the partition of the urinary sulphur of calves during feeding and during starvation*

Fraction of sulphur	Preliminary period (mg./day)	Starvation period (mg./day)	Increase (mg./day)
Inorganic	210·4	382·2	+171·8
Ethereal	66·3	63·3	−3·0
Neutral	93·8	95·1	+1·3
Total	370·5	540·6	+170·1

The constancy of the neutral S fraction is in agreement with the contention that it is an endogenous fraction, though the experiments of Amann (1933) suggest that its excretion is by no means constant, increasing markedly when very high protein diets are given. The constancy of the ethereal-sulphate fraction was not expected. It was thought that there would be a reduction in the quantity of phenols formed by putrefaction and stasis in the gut on starvation with a consequent decline in the sulphate-ester excretion. Possibly the continuing excretion of faeces during fasting indicates that there are present in the intestinal tract sufficient food residues to give rise to phenols in comparatively large quantities.

The ratio N:S in the urine of man during starvation varies between 15 and 17 (Lusk, 1928; Benedict, 1915; Cathcart & Green, 1913). The N:S ratio in skeletal muscle is 13·4 (Wilson, 1925). In the calf the N:S ratios are comparable to those found in starvation metabolism in man but are higher than would be expected if muscle proteins and their constituent S-containing amino-acids were the source of both the N and the S. The explanation probably is that neither the total S nor the total N entirely originates in muscle katabolism (see Table 12 and subsequent discussion), a small part in each instance coming from the so-called endogenous metabolism. If it is assumed that the excretion of neutral S is a measure of a minimal S excretion, and the excretion of N above the endogenous level is taken as representing katabolism of body protein, the ratio of non-endogenous N to non-endogenous S is 13·8, a value in fair agreement with Wilson's value for the N:S ratio in muscle.

It may be concluded, therefore, that there is an increase in S excretion during fasting following a milk-protein diet, the increase being entirely in the inorganic fraction, and that, subject to one assumption, the excretion of S is compatible with the hypothesis that body protein is being katabolized in large amounts.

Excretion of acetone in the urine

The daily excretion of acetone is shown in Fig. 3, and the mean results are summarized in Table 17. It is clear from Fig. 3 and Table 17 that no ketosis occurred on starving the young calf. This agrees with the results obtained in the mature ruminant by Sjollema & van der Zande (1923), Carpenter (1927) and Hutchinson & Morris (1936*b*),

none of whom observed any ketosis in cattle on starvation. This result in the young calf, which is to all intents a non-ruminant, simple stomached animal, is of considerable interest, for in man many g. of ketone bodies are excreted during fasting. The extent of the ketosis is much smaller in the poorly nourished individual (Deuel & Gulick, 1932), suggesting that in the calf, an animal possessing only small fat reserves, ketosis would not be a symptom of starvation. In this respect, however, infants and children evidently develop a more intense ketosis on starvation than adults (Gamble, Ross & Tisdall, 1923).

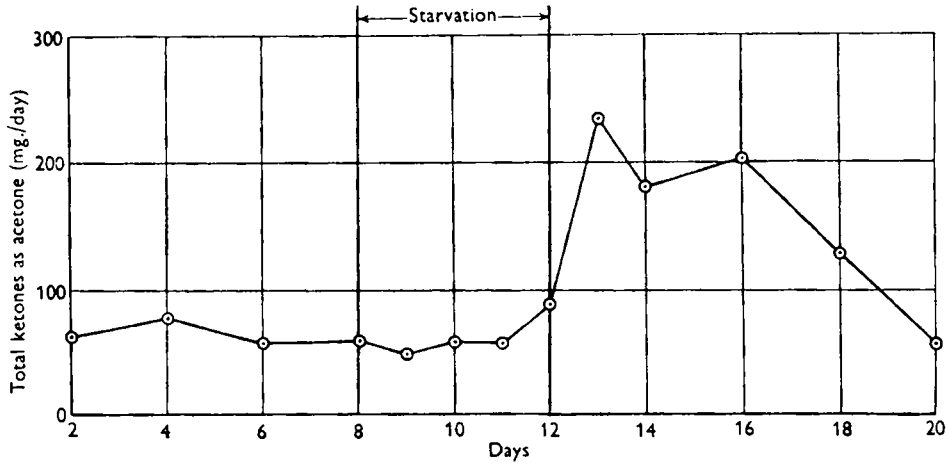

Fig. 3. Effect of starvation on the urinary excretion by calves of ketone bodies.

Table 17. *Daily excretion of ketone bodies in the urine of the calves during feeding and during starvation, expressed as acetone in mg./day*

Period	Calf no. 4 Level of feeding		Calf no. 5 Normal level of feeding	Mean
	Normal	Low		
Preliminary	60·7	58·5	68·7	62·6
Starvation	49·4	54·3	89·2	64·3
Recovery	95·7	134·2	120·3	116·7

It is of interest that an increase in urinary acetone excretion occurred on realimentation. This suggests that there is a slight disturbance of carbohydrate metabolism at this time. It may be suggested that the large demand for glucose to replenish the depleted glycogen resources of the animal results in an increase in fatty-acid oxidation following fasting.

Excretion of minerals in the urine

As already stated, calcium, magnesium, sodium, potassium, chlorine and phosphorus were determined on 2-day samples of urine collected during the three periods of each experiment. The results are shown in Table 18. Balances were not determined, and it must be remembered that the sampling errors involved in obtaining these results may have been high.

Table 18. *Daily excretion of mineral elements in the urine of the calves during feeding and during starvation (mg.)*

Calf no.	Level of feeding	Period	Cl	P	Na	K	Mg	Ca
4	Normal	Preliminary	2467	227	914	2678	62	149
		Starvation	664	312	661	1493	34	75
		Recovery	2884	399	1398	2780	30	106
4	Low	Preliminary	2003	245	529	2027	13	135
		Starvation	274	143	330	1569	30	33
		Recovery	2614	345	876	3291	18	87
5	Normal	Preliminary	3057	182	338	3145	45	94
		Starvation	140	184	—	1315	8	22
		Recovery	2379	28	313	1496	18	71
Mean loss/day during starvation			326	213	495	1456	24	43

It is clear that there was a reduction in the excretion of Cl, K, Na and Ca during starvation and no constant change in the other elements. The mean daily loss during starvation may be used to indicate the extent of the katabolism. If only muscle tissue was being broken down, and if the K loss is assumed to have come entirely from muscle, one can estimate the approximate excretion of other elements which could have come from the muscle substance. This can only be an approximation, for the complete analytical figures for muscle are very few and were mostly obtained many years ago, whereas the urinary excretion as determined is subject to a fairly large sampling error. By use of ratios, however, part of the sampling error is minimized. The final results of the calculation are shown in Table 19.

Table 19. *Calculation of approximate amounts of body protein katabolized, based on the analysis of muscular tissue and on urinary excretion of the calves*

Element	Amount in muscle when 1500 mg. K are present* (mg.)	Mean daily excretion in the urine (mg.)	Conclusion
K	1500	1456	—
Na	400	495	Loss of extracellular fluid
Cl	350	326	—
Ca	35	43	No loss of minerals from bone
Mg	105	24	Retention of essential enzyme systems of the cell
P	750	213	

* See text above.

It would appear from Table 19 that there was no katabolism of bone minerals during starvation. This is not in agreement with some of the data obtained on human beings (Peters & Van Slyke, 1931) but, as has been pointed out, part of the bone loss in man may be due to the accompanying acidosis. Hawk, Oser & Summerson (1947), however, point out that long periods of starvation in the dog (up to 104 days) do not cause any marked loss of minerals from the bone.

The larger quantity of Na present in the urine suggests a loss of extracellular fluid

during starvation. The smaller quantities of Mg and P present in starvation suggest that there was no extensive loss of nucleoprotein material and that the enzyme systems of the cell remain intact. This is in substantial agreement with the results previously reported on the absence of an increase in purine katabolism during a fast.

Energy metabolism and respiratory exchange

Accuracy of the determinations. In that the interpretation of the results of the respiratory-exchange determinations depends largely on the accuracy with which the determinations were made it is essential to have information on the variation associated with each determination. It may be stated at the outset that the method

Table 20. *Regression equations relating functions measured during determinations of starvation respiratory exchange to length of starvation, with coefficients of variation estimated by analysis of variance of the regression*

Function	Equation	Percentage decline/ day	Variance ratio (e^{z_2})	Coefficient of variation (%)
Carbon-dioxide production (l./hr.)	$x = 10{\cdot}117 - 0{\cdot}600D$	5·93	736·4	1·4
Oxygen consumption (l./hr.)	$x = 12{\cdot}997 - 0{\cdot}755D$	5·81	233·2	2·0
R.Q.	$x = 0{\cdot}784 - 0{\cdot}006D$	7·96	3·7	1·8
Heat production (Cal./hr.)	$x = 61{\cdot}67 - 3{\cdot}76D$	6·10	502·4	1·4
Pulse rate (beats/min.)	$x = 79{\cdot}7 - 6{\cdot}38D$	8·00	498·3	2·0
Respiratory rate (respirations/min.)	$x = 16{\cdot}6 - 1{\cdot}15D$	6·93	7·0	13·9
Minute volume of respiration (l.)	$x = 4{\cdot}40 - 0{\cdot}032D$	7·36	17·8	13·8
Body-weight (kg.)	$x = 35{\cdot}88 - 0{\cdot}655D$	1·83	14·9	1·6

D: days of starvation.

adopted for determination of respiratory exchange was extremely sensitive. Slight head movements of the calf in a duplicate run invariably could be detected in a higher consumption of oxygen and production of carbon dioxide. The accuracy of the method was determined by analysis of variance, computing the coefficient of variation from the mean and the standard deviation of the residuals from a fitted linear regression. The results for calf no. 4, which are typical, are shown in Table 20. From the results in that table it is clear that the errors attached to the determinations were very small. For oxygen consumption, heat production and carbon-dioxide production the errors expressed as a percentage of the mean are all less than 2. The respiratory rate and ventilation rate per min. were slightly more variable, but even so were well within the range of variability one might expect in a function under partial voluntary control.

The course of heat production during fasting with reference to the constancy of the basal metabolism of the calf. From Table 20 it is clear that the linear component of the regression was very highly significant in calf no. 4 and that the residual variance was very small. In both calves heat production following feeding declined slowly over the whole 4 days of observation. The same was true of each function studied, pulse rate, respiratory rate and minute volume. The data relating to each animal are shown in Table 21.

From the equations in Table 21 it can be seen that both calves showed a marked

decline in heat production throughout the fasting period. There was no indication at any time that a constant level of metabolism had been reached, and the regression equations were not in any respect non-linear, as can in fact be inferred from the errors shown in Table 20. In every instance the decreases in oxygen consumption, heat production and body-weight with continued fasting were all highly significant, P being always smaller than 0·01 and sometimes smaller than 0·001.

Table 21. *Regression equations showing the fall in heat production of the calves with continued starvation, the fall in oxygen consumption and the decline in body-weight*

Calf no.	Level of feeding	Function	Regression equation	Daily percentage decline
4	Normal	Oxygen consumption (l./hr.)	$x = 13{\cdot}00 - 0{\cdot}600D$	5·81
		Heat production (Cal./hr.)	$x = 61{\cdot}67 - 3{\cdot}76D$	6·10
		Body-weight (kg.)	$x = 35{\cdot}88 - 0{\cdot}655D$	1·83
4	Low	Oxygen consumption (l./hr.)	$x = 13{\cdot}28 - 0{\cdot}454D$	3·42
		Heat production (Cal./hr.)	$x = 53{\cdot}67 - 3{\cdot}32D$	6·19
		Body-weight (kg.)	$x = 38{\cdot}83 - 0{\cdot}525D$	1·35
5	Normal	Oxygen consumption (l./hr.)	$x = 14{\cdot}86 - 0{\cdot}68D$	4·58
		Heat production (Cal./hr.)	$x = 64{\cdot}58 - 4{\cdot}36D$	6·76
		Body-weight (kg.)	$x = 38{\cdot}89 - 0{\cdot}315D$	0·81

D: days of starvation.

The heat production was computed using the mean excretion of N/day. The rate of decline in heat production was much greater than the decline in body-weight, which means that, not only the metabolism of the animal per kg. body-weight declines markedly during fasting, but that its metabolism per unit of surface area declines at an even greater rate, in that surface area tends to be proportional to a fractional power of body-weight between 0·6 and 0·8. It will be noted that the rate of decline of heat production was the same whether or not calf no. 4 had received an adequate or a reduced amount of diet, and that the rate of fall in the heat production of calf no. 5 was also similar.

It is clear that this fall in heat production of calves during starvation is not only highly significant in the single individual but is reproducible as between individuals and not affected by the earlier nutrition of the individual within the limits employed. These results are in marked contradistinction to those found with man and mature animals. In the first place, though in man there is a fall in total heat production in long-continued fasting, fasting for 1 week does not result in a large fall in metabolism per kg. body-weight (Lusk, 1928; Benedict, 1907, 1915; Lehmann & Zuntz, 1893). In the cow, the metabolism/kg. body-weight falls during the 1st day of starvation, largely owing to the long period needed to reach a postabsorptive state. The metabolism of the animal does not appear to decline markedly once this level has been attained (Benedict & Ritzman, 1927). In sheep, the observations of Blaxter (1948) and Marston (1948) indicate that a plateau in metabolism occurs. Prolonged starvation in the rat, however, reduces heat production whether expressed per kg. body-weight or per sq.m. body surface (Benedict & Fox, 1934), and in mice a marked fall in metabolism, as judged by body-temperature changes, occurs during continued fasting (Buschke & Vasarhelyi, 1932).

Data relating to this decline in heat production in other species are given in Table 22. Statistical analyses were made of the published results by the methods used in dealing with the calf results.

Table 22. *Regression of heat production (Cal./kg. body-weight) on length of fast in days; a comparison of the calf with man and the steer*

Species	Reference	Regression equation	Length of observation (days)	Statistical significance
Calf	Present results	Cal. = 37·03−1·98*D*	4	Very highly significant
Man	Benedict (1907)	Cal. = 32·28−0·66*D*	5	N.S.
Man	Lusk (1928)	Cal. = 30·97−0·39*D*	6	N.S.
Steer C	Benedict & Ritzman (1927)	Cal. = 16·62−0·47*D*	6	N.S.
D		Cal. = 17·12−0·58*D*	6	N.S.
E		Cal. = 22·4 −0·04*D*	4	N.S.

D: days of starvation. N.S.: not significant.

The second aspect of this decline in heat production following fasting is that it is not affected to any appreciable extent by the earlier nutritional level of the individual. There is a reduction of the intercept when $D=0$ (Table 22), but no change in the percentage decline. Marston (1948) has shown that heat production of sheep fed on a high plane of nutrition fell to a constant basal level more slowly (over a period of 7 days) than the heat production of animals fed on a submaintenance diet which reached constancy after only 2 days of fasting. Marston has attributed this difference in the shape of the curve of heat production during fasting to the failure of the animals given the higher level of energy to reach a postabsorptive state.

The reason for this decline in metabolism in prolonged fasting in the calf is not clear. The most logical supposition would be that a postabsorptive state had not been reached in the calf starved for 4 days. This is not supported by the results for the following reasons: (1) The respiratory quotient did not show any marked decline during the fast. This was also true of the non-protein R.Q. (2) The peak of creatine excretion in the urine occurred early in the starvation period, indicating that the animal was very close to a postabsorptive state even on the 1st day of starvation. (3) There was no significant difference in the rate of decline in metabolism following different levels of food intake, as has been shown to occur in the sheep. (4) The fall in metabolism was not exponential and the experimental points did not deviate from a linear rate of decline. If the decline in metabolism was due to the metabolism of food residues an asymptote would be expected.

The decline in metabolism was thus not due to failure to reach the postabsorptive state. This is reasonable in so far as the rat reaches a basal level of metabolism at about 14 hr. (Wesson, 1930–1) and man at about 12 hr. It is only herbivora that take 72 hr., or more.

A likely explanation is that there is a marked reduction in muscular tone and in the small, almost involuntary, skin and muscle movements during prolonged fasting in the calf. Body temperatures were unfortunately only recorded on one occasion, when, at

the end of fasting, a value of 99·4° F. was obtained, compared with a previous value of 101·5° F. at the low level of feeding.

Relation between the heat production of the calf and its cardio-respiratory activity. As heat production fell, the pulse rate, respiratory rate and minute volume of the respiration also fell in both the calves. That the pulse rates of sheep and cattle are approximately proportional to their metabolism is well known and has recently been studied by Blaxter (1948). Fig. 4 shows the relationship in calf no. 4, with the results pooled for both periods of starvation. The equation relating metabolism to pulse rate was:

$$\text{Cal./hr.} = 10{\cdot}46 + 0{\cdot}644 \times \text{pulse rate.}$$

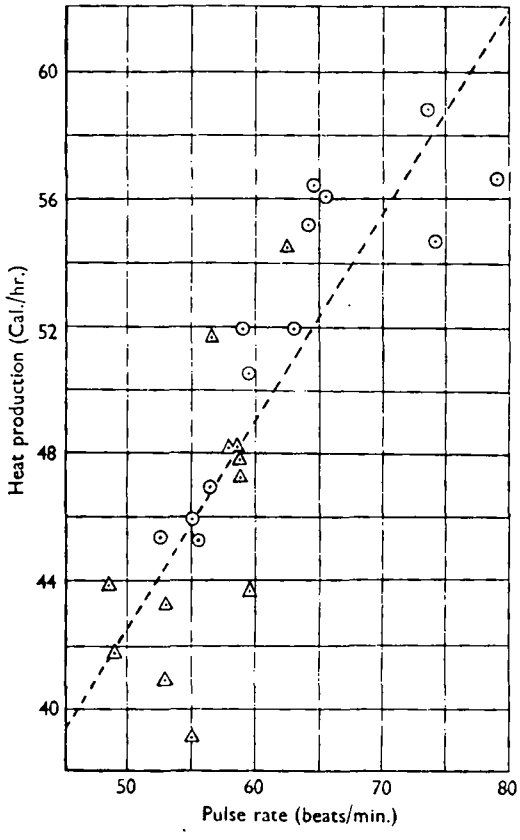

Fig. 4. Relation between pulse rate and heat production in calf no. 4. ⊙, first starvation experiment; △, second starvation experiment.

This regression was very highly significant statistically. By calculating this equation to the basis of Cal./sq.m. body surface/24 hr. it may be compared with similar equations computed for the sheep and the steer. The transformed equation is shown in Table 23.

It would appear that the relation between pulse rate and metabolism is fairly similar in the young calf and the adult sheep, animals of about the same size, but that in the steer the heat production is proportionately much greater per unit of body surface for comparable pulse rates.

Table 23. *Relation between heat production and pulse rate in the calf, the sheep and the steer*

Species	Weight (kg.)	Equation	Heat production (Cal./sq.m. body surface/24 hr.), with pulse rate beats/min. 50	70	90
Calf*	40	$M=13{\cdot}6P+220{\cdot}0$	900	1172	1444
Sheep	40	$M=17{\cdot}2P-117{\cdot}5$	743	1086	1431
Steer	400	$M=16{\cdot}4P+831{\cdot}4$	1650	1978	2306

* Surface area taken to be 1·14 sq.m.

P: pulse rate (beats/min.). M: heat production (Cal./sq.m. body surface/24 hr.).

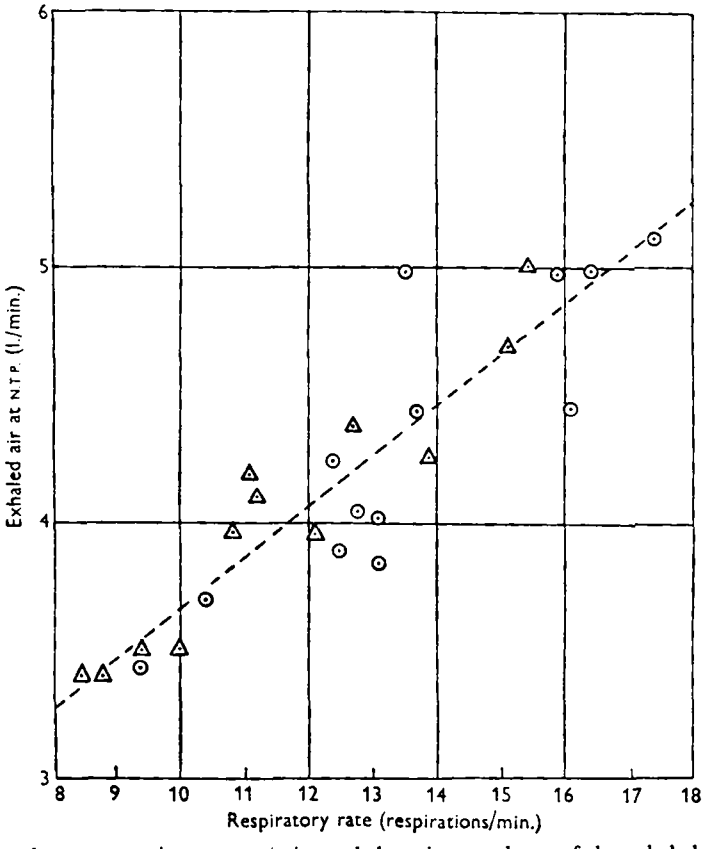

Fig. 5. Relation between respiratory rate/min. and the minute volume of the exhaled air in calf no. 4. ⊙, first starvation experiment; △, second starvation experiment.

The relation of the minute volume to the respiratory rate of the calf during fasting is shown in Fig. 5. From this it is clear that an increase in respiratory frequency is associated with an increase in minute volume. This is not a direct proportionality, for the tidal air of the respiration declines with increasing respiratory rate as shown in Fig. 6. The equation for the straight line regression is:

$$\text{Tidal air, in ml. at N.T.P.} = 482{\cdot}4 - 11{\cdot}75 \times \text{respiration rate.}$$

This equation applies only within the limits of 7–20 respirations/min. and within this

range may be regarded as highly significant statistically. In the sheep, an animal of closely similar size, the equation relating tidal air to respiratory frequency has a regression coefficient of only 1·9, compared with 11·8 in the calf. The sheep experiments were all associated with respiratory rates greater than 25/min., even during prolonged fasting, and the two sets of observations are probably not fully comparable (Blaxter, 1948).

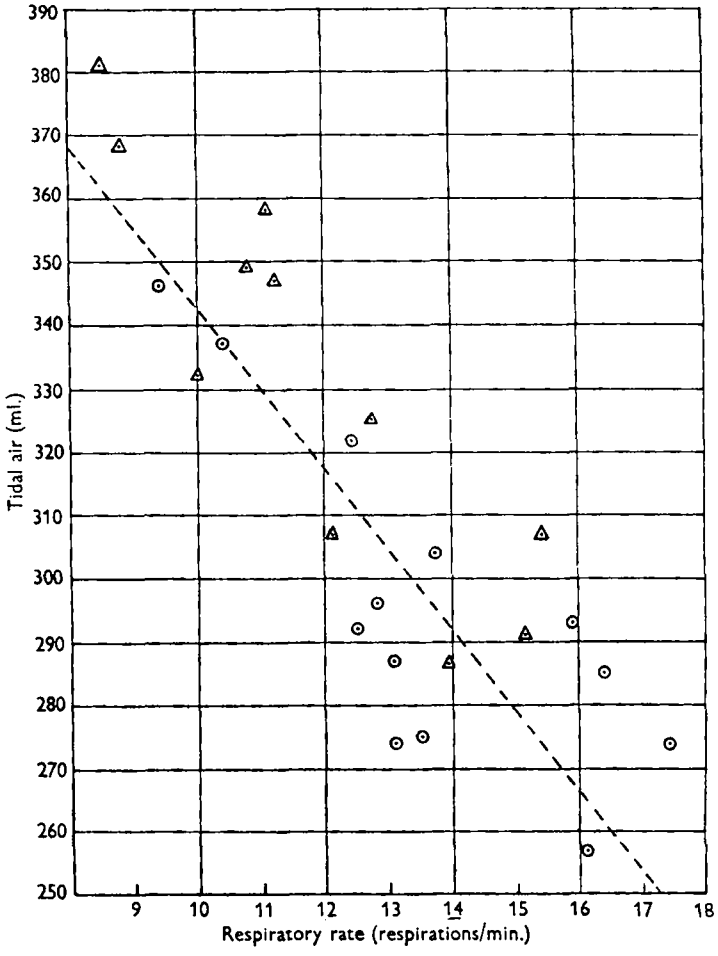

Fig. 6. Relation between respiratory rate/min. and tidal air in calf no. 4. ⊙, first starvation experiment; △, second starvation experiment.

DISCUSSION

Materials katabolized during fasting in the calf, and the loss in body-weight

The respiratory quotient of an animal when corrected for total urinary N excretion using the factors of Loewy (1911) or of Kriss & Miller (1934) may be used to compute the proportions of fat and carbohydrate oxidized in the body. This method has recently fallen into disrepute (Soskin & Levine, 1946) largely owing to the realization that reactions other than simple total oxidation of fat and carbohydrate are always occurring. As pointed out by Dewar & Newton (1948–9), over long periods of time

this criticism is not valid, for the intermediate side-reactions resulting in abnormal respiratory quotient will be cancelled out. In long-term experiments carried out over 24 hr. periods, the R.Q. gives substantially the proportion of energy derived from the ultimate oxidation of fat and carbohydrate. In the present experiments the experimental periods were only 45 min. in total duration (including preliminary periods) and 'abnormal' R.Q.'s might therefore be expected. In the first experiment with calf. no. 4, the non-protein R.Q. remained high throughout, indicating that in the early stages of the fast 10–12 % of the heat production was arising from complete oxidation of carbohydrate. In the remaining experiments non-protein R.Q.'s were quite normal, varying within small limits. On occasions, however, values below 0·707 were obtained, indicating either that complete collection of the carbon dioxide produced was not obtained owing to underventilation or that momentarily the determination had coincided with a phase of intermediary metabolism characterized by a low apparent non-protein R.Q. In no instance was the extremely wide variation of R.Q. met by Werthessen (1937) or Markowitz (1946) ever observed, any variation from the general trend of the non-protein R.Q. with time being within limits of $\pm 0{\cdot}05$.

It would appear therefore that the present results may be used to compute the fat and carbohydrate oxidized or dissimilated, and for the reason given above, the results for calf no. 4, period 1, have been excluded as far as a partition of the non-protein heat production is concerned. Table 24 summarizes the results of the calculation of the

Table 24. *Estimate of constituents lost daily from the body of the calf during starvation*; *calf no.* 4, *period* 2 *and calf no.* 5, *period* 1

Constituent of body	Daily loss (g.)
Body-weight	525
Body fat	99
Body protein	54
Body carbohydrate	13
Extracellular water	39
Intracellular water	326
Total weight loss accounted for	531

materials katabolized. The extracellular and the intracellular water losses are based on the molal quantities of sodium and potassium excreted in the urine during fasting and calculated using the equations of Gamble *et al.* (1923). The table shows that by far the greatest loss to the body is water and that most of this is from the cells rather than from the intercellular spaces. The loss of extracellular water is small and indicates only a slight dehydration of the animals. The loss of body fat of nearly 100 g./day is the mean loss on the 2nd day of starvation. Towards the end of the starvation period, on the 4th day, proportionately far more protein (up to 24 % of the total calories in calf no. 4) was lost to the body. In this respect, it has to be remembered that not the whole of the urinary N is necessarily a reflexion of protein katabolism. This has been adequately discussed in the section dealing with urine composition (p. 39), and serves to indicate that the protein value in Table 24 is probably too high an estimate, part of the loss of N being of body non-protein nitrogen constituents.

We have not determined the source of the katabolized protein, fat and carbohydrate, but have shown that there is no important loss of nucleic-acid derivatives on fasting the calf. Kosterlitz & Campbell (1945) have recently reviewed aspects of the loss of N-containing compounds from the body in starvation and have pointed out the relatively large loss of N from the liver of the fasting animal. This loss is accompanied by a loss of ribonucleic acid from the liver cytoplasm (Kosterlitz, 1944; Davidson & Waymouth, 1944*a*, *b*). There is no reason to believe, on the basis of chemical analysis, that in the loss of muscle substance during starvation nucleic acid and protein are not lost simultaneously. On the other hand, in muscle in starvation there is both an absolute and a relative increase in the number of cell nuclei (Roche & Hoerner, 1933), and in the liver only the labile liver cytoplasm is reduced (Kosterlitz, 1944; Campbell & Kosterlitz, 1947) while the number of cell nuclei with their large store of deoxyribonucleic acid remains intact (Davidson & Waymouth, 1944*a*; Davidson, 1945).

In the calf there was no indication of an acceleration of purine excretion during starvation, indicating that adenine and guanine were not arising from cytoplasmic ribonucleic acid or nuclear deoxyribonucleic acid in large quantities. This is compatible with the contention that the major portion of the N loss was from the non-nuclear portion of the muscles, with relatively very much smaller portions arising from the liver.

Fasting the calf as a method for the control of diarrhoea

From the above discussion it is clear that fasting the young calf causes marked losses of its body protein and fat, and that, because its metabolism is more intense than that of the older animal, its losses are proportionately more severe (see Tables 9, 11 and 22). A higher percentage of the total heat loss of the calf comes from the degradation of body protein than in the mature animal. This is in complete accord with the classical work of Voit (1901) who showed that the quantity of the protein metabolism in starvation depends upon the amount of fat in the body. The calf has little body fat at birth or in the early stages of growth (Armsby & Moulton, 1925), and the effect of starvation is therefore intense. If calves are affected by diarrhoea, and fasting is used as a method of control, it follows a period in which depletion of reserves has already occurred. Realimentation after starvation, if too rapid, may lead to an exacerbation of symptoms, as shown by the behaviour of calf no. 5 during the first period of starvation. Lastly there is no indication that, following realimentation, efficiency of food utilization is enhanced, and the loss in weight, body protein, and body fat can only be restored by establishing a plane of nutrition higher than that before fasting was commenced.

SUMMARY

1. Experiments are described in which two young calves were fasted for periods of 4 days and their metabolism of nitrogen, energy, chloride, potassium, sodium, magnesium, calcium and phosphorus was determined.

2. The mean loss of weight on fasting was 578 g./day. Following the fast, gain in weight, N storage and digestibility of the diet were the same as in the period before the fast.

3. Faeces were produced during fasting in smaller amounts, but of the same dry-matter composition.

4. The loss of urinary N during starvation averaged 250 mg./kg. body-weight/day, a value much greater than in either mature ruminants of the same body size or of the same species.

5. The biological value of the protein was slightly reduced following a fast, but the reduction was not statistically significant. At both levels of feeding, dietary energy supply rather than protein supply was the factor limiting growth.

6. The excretion of urinary metabolites containing N showed that marked increases occurred only in the urea, creatine and uric-acid fractions. Of the increase in excretion over the endogenous metabolism, 94 % was accounted for by excretion of urea, ammonia and creatine. There was no increase in the total excretion of purine N, the small increase in uric acid being compensated for by a fall in allantoin excretion. The distribution of urinary N of the calf during starvation differs from that of the cow, goat or sheep largely because of the increase in the katabolism of body protein.

7. Urinary excretion of sulphur increased, the increase being all in the inorganic fraction, neither the ethereal nor neutral S changing. The ratio N:S of the increased metabolism over the endogenous metabolism indicated that the source of the S was body-muscle protein.

8. There was no acidosis or ketonuria during starvation of the calves. A slight ketosis during realimentation was a constant symptom.

9. Urinary Cl, K, Na and Ca fell during starvation. There was no evidence of bone katabolism during fasting, and the mineral loss could be traced to katabolism of the cell. The losses of Mg and P were smaller than could be accounted for by complete breakdown of the cell, suggesting that nuclear material was not being katabolized in any large quantities.

10. The respiratory exchange was characterized by a constant fall in metabolism throughout the 4 days of the fast. This fall was not affected by the plane of nutrition of the calves, and was three times greater than the slow fall of metabolism that occurs in man with continued fasting. Evidence is produced to show that this fall was not due to failure to reach a postabsorptive state. The fall was at a much greater rate than the weight loss of the calves.

11. The pulse rate fell during starvation and an equation for estimating metabolism from pulse rate is presented. The relation between respiratory rate and minute volume of the respiration is discussed.

12. The loss of weight during fasting was quantitatively accounted for as fat, protein and carbohydrate katabolized and as loss of water from extracellular and intracellular compartments.

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The Nutrition of the Young Ayrshire Calf

4. Some Factors Affecting the Biological Value of Protein Determined by Nitrogen-Balance Methods

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The biological value of a protein as evaluated by the Thomas-Mitchell procedure (Mitchell, 1923–4) is affected to a considerable extent by the percentage of protein in the diet (Mitchell, 1923–4; Mitchell & Beadles, 1926–7; Hamilton, 1938). When the percentage is high, a larger proportion of the absorbed amino-acids is deaminated, the nitrogen appearing in the urine and the non-nitrogenous moiety ultimately being assimilated as a source of energy. It is only when the protein content of the diet is such that the demand by the tissues for amino-acids is greater than the supply, that maximal biological values are obtained. This inevitably entails partial protein deficiency in the animal and a submaximal rate of growth.

In the diet of the young growing rat the percentage of protein usually adopted for the determination of biological values is 8, whereas in adults percentages as low as 4–5 have to be used to ensure that tissue demand for amino-acids is greater than supply. Similar percentages were adopted in experiments with growing sheep and with growing cattle (Miller & Morrison, 1939, 1942; Harris & Mitchell, 1941; Swanson & Herman, 1943). Experiments designed to study the biological value of dietary protein in the calf during the first few weeks of life do not appear to have been conducted, and