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**THE FEEDING VALUE OF EARLY CUT AND
LATE CUT HAY FOR THE MAINTENANCE
OF COWS**

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CHAPTER I

INTRODUCTION

Hay of mixed grasses is the main winter feed of cattle in the Netherlands. Therefore, a good understanding of its nutritive value is of primary importance. This value varies however a good deal according to the stage of growth of the grass, weather conditions during hay curing, methods of hay making, heating in the hay stack, kind of animal production etc. Therefore, extensive investigations are necessary for obtaining a good understanding of the importance of all these factors.

Digestion trials with various kinds of hay have been performed in the Netherlands many years ago (BROUWER and DIJKSTRA, 1938) and have later on largely been extended by DIJKSTRA so that at present extensive information on this subject is available.

The feeding value of roughages, however, depends not on digestible constituents only. KELLNER (1900) showed already in difference trials that the efficiency of the digestible energy for fat production varies with the crude fibre content of the ration. He also showed (1911) that the efficiency of the digestible energy for milk production is higher than for lipogenesis. MØLLGAARD's calculations (1929, 1941) based on data in the literature, suggested a higher efficiency for maintenance than for synthesis of body fat.

As roughages are almost the only components in the cow's rations for maintenance and for use in periods of low production it was thought appropriate to start with determination of the feed value of hay for maintenance only. In this way early and late cut hay from four fields were examined.

Originally it was thought that for these determinations of maintenance value difference trials with two or three periods might not be necessary but that a number of experiments consisting of one single period might be feasible just as in the determination of digestibility. It was however shown (VAN ES, 1961) that even in experiments performed with utmost care, variations in maintenance requirement are rather large. Therefore, the early and late cut hay from each field were always examined simultaneously with the reversal method and with the use of two identical respiration chambers.

Methods and Experimental procedures are described in chapter II and III. The main results of the trials have been calculated in two ways: firstly by Regression analysis (chapter IV) and secondly by Analysis with corrections of energy balances from literature (chapter V). After that a Discussion and Summary follow.

CHAPTER II

METHODS

Experiments were conducted with non-lactating non-pregnant Friesian cows fed on four sets of early and late cut hay. The hay of each set came from alternate strips of one pasture. So much hay was given that the animals were approximately in energy equilibrium. Each set was usually fed to six cows in reversal trials with double periods.

There were nine double periods: R 17/18, R 19/20, R 25/27, R 29/30 etc. In most double periods there was performed one reversal trial, consisting of two double experiments with one cow each. In the first single period of such a double period one animal was given the early cut hay and its companion the late cut hay and in the next single period the rations were reversed. One reversal trial consisted therefore of a block of four single experiments; i.e. two double experiments with one cow each. In some double periods not one but two reversal trials were conducted simultaneously. The total number of single experiments was 44. A survey of the experiments is given in table II.1. The design of one double period with two reversal trials is as follows.

| | | First reversal trial | | Second reversal trial | |
|-----------------------|--------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| | | Double exp. with cow 1 | Double exp. with cow 2 | Double exp. with cow 3 | Double exp. with cow 4 |
| Double period R 20/21 | Single period R 20 | Single exp. with ration A | Single exp. with ration B | Single exp. with ration A | Single exp. with ration B |
| | Single period R 21 | Single exp. with ration B | Single exp. with ration A | Single exp. with ration B | Single exp. with ration A |

Each single period and each single experiment covered a preliminary period and an experimental period, the latter subdivided in subperiod I and subperiod II.

II.1. ANIMALS

The nine Friesian cows used in the experiments were adult, non-pregnant, non-lactating and healthy. They were obtained from the experimental farm of the laboratory. In selecting them no special attention was paid to their previous levels of milk production or to their condition. All animals were accustomed to

TABLE II.1. Number, month and year of single period; body weight of the animals

| Animal | 1958 | | 1959 | | 1960 I | | 1960 II | |
|---------------|------------------------|------------------------|-----------------------|-----------------------|------------------------|------------------------|-----------------------|-----------------------|
| | Early | Late | Early | Late | Early | Late | Early | Late |
| Zwartschoft 1 | R 17 11-'58 598 kg | R 18 12-'58 619 kg | R 25 12-'59 561 kg | R 27 2-'60 600 kg | R 33 10-'60 604 kg | R 34 11-'60 613 kg | R 35 12-'60 597 kg | R 36 1-'61 602 kg |
| Zwartkop | R 18 12-'58 521 kg | R 17 11-'58 549 kg | R 27 2-'60 619 kg | R 25 12-'59 611 kg | R 34 11-'60 566 kg | R 33 10-'60 576 kg | R 36 1-'61 567 kg | R 35 12-'60 571 kg |
| Alke | R 20 2/3-'59 488 kg | R 19 1/2-'59 516 kg | R 29 4-'60 566 kg | R 30 5-'60 567 kg | R 34 11-'60 551 kg | R 33 10-'60 585 kg | R 36 1-'61 558 kg | R 35 12-'60 584 kg |
| Kee 2 | R 19 1/2-'59 540 kg | R 20 2/3-'59 561 kg | R 29 4-'60 566 kg | R 30 5-'60 567 kg | R 33 10-'60 529 kg | R 34 11-'60 561 kg | R 35 12-'60 541 kg | R 36 1-'61 548 kg |
| R. Willy | R 31 6-'60 562 kg | R 32 7/8-'60 591 kg | R 30 5-'60 562 kg | R 29 4-'60 575 kg | R 38 3/4-'61 566 kg | R 39 4/5-'61 587 kg | R 40 6-'61 568 kg | R 41 7-'61 572 kg |
| Zwartschoft 2 | R 32 7/8-'60 559 kg | R 31 6-'60 582 kg | | | R 39 4/5-'61 545 kg | R 38 3/4-'61 594 kg | R 41 7-'61 541 kg | R 40 6-'61 573 kg |
| Kee 3 | | | | | | | | |
| Jansje | | | | | | | | |
| Roosje | | | | | | | | |

wear a harness with urinal for separate collection of faeces and urine and to stay in the respiration chambers before each experimental period started. Some data on the animals (birthday, age, previous milk production) are given in table II.2.

In period R 27 cow Kee 2 and in R 29 the cows Zwartschoft 2 and Roorda Willy had mastitis in the first part of the preliminary period. Treatment with penicillin and terramycin gave a quick recovery. The preliminary period of the experiment with Kee 2 was prolonged with four days. The figures for maintenance requirement ($M_{m,500}$) of Kee 2 and Zwartschoft 2 are rather high in the experimental period succeeding this preliminary period.

Zwartschoft 2 lost appetite and had a high body temperature in the second week of the preliminary period of R 31 but recovered after a few days. Nevertheless, this preliminary period also was prolonged with some days.

At the time that they were used first the animals were in a good condition, neither meagre nor fat.

Most cows remained in this condition during the experiments, but two of them, Alke and Kee 2, gained considerably between the periods R 20 and R 25, mostly while on pasture in summer.

Cow Jansje gradually developed a bull behaviour in the course of the experiments. Autopsy showed cystic ovaria and enlarged adrenals.

II.2. PRELIMINARY AND EXPERIMENTAL PERIODS

It has already been mentioned that there were nine double periods each consisting of two single periods succeeding each other closely. Each single period consisted of a preliminary (or a transition) period and an experimental period. In each double period one or two reversal trials were run, each consisting of a block of four single experiments, according to the design mentioned above.

Each single experiment lasted five weeks. It consisted of two parts, covering the preliminary period and experimental period respectively. In all periods in which two animals were used at the same time (period R 17–32, R 38–41) the experimental period lasted 14 days. In the periods with four animals (R 33–36) it lasted two days longer. The experimental periods were subdivided in a subperiod I and a subperiod II.

During the last week of the preliminary period, during the experimental period and for two days following this period the same amount of the same feed was fed daily. Except for the first part of the preliminary period all rations were weighed on one day. Approximately half of the day ration was given at 16.45, the remainder at 7.00 next morning, but during the first two weeks of the preliminary period at 8.00.

During the experimental period faeces and urine were daily collected, weighed

TABLE II.2. Age and milk production of the animals

| Name | Birthday | Age in years during period | Lactation | | Milk produced (kg) | Milkfat produced (kg) |
|--------------|----------|----------------------------|---------------------|--------|--------------------|-----------------------|
| | | | Nr. | Length | | |
| | | | | Year | | |
| Zwartshoof 1 | 18-6-'50 | R 17-18 : 8½ | 4th | 419 d. | 4437 | 161 |
| | | | 5th | 423 d. | 5795 | 203 |
| Zwartkop 2 | 29-3-'51 | R 17-18 : 7½ | 4th | 319 d. | 4610 | 191 |
| | | | 5th | 242 d. | 3894 | 145 |
| Alke | 5-'52 | R 19-20 : 7 | 3rd | 289 d. | 4321 | 147 |
| | | | 4th | 264 d. | 4245 | 136 |
| Kee 2 | 11-'50 | R 19-20 : 8 | 4th | 399 d. | 5343 | 214 |
| | | | 5th | 305 d. | 4660 | 181 |
| R. Willy | 20-3-'51 | R 29-32 : 9 | 4th | 291 d. | 4129 | 168 |
| | | | 5th | 299 d. | 3854 | 155 |
| Zwartshoof 2 | 26-9-'51 | R 29-32 : 8½ | 4th | 295 d. | 5000 | 185 |
| | | | 5th | 310 d. | 4870 | 177 |
| Kee 3 | 21-2-'57 | R 33-36 : 4 | one incomplete list | | | |
| | | | 3rd | 371 d. | 4114 | 146 |
| Jansje | 19-4-'54 | R 38-41 : 7 | 4th | 245 d. | 4128 | 149 |
| | | | 3rd | 249 d. | 3015 | 120 |
| Roosje | 8-1-'54 | R 38-41 : 7½ | 4th | 175 d. | 2704 | 116 |

and sampled. The collection of urine started at 9.00 and the collection of faeces the next morning again at 9.00.

Originally four, but after R 20 six respiration trials of 24 hours each were conducted in the single experiments.

During the experimental period and, if necessary, earlier the rectal temperature of the animals was measured daily just before the afternoon feeding except when the animals were in the respiration chambers.

In the experimental period the animals were weighed three or four times, usually at about 11.00 but occasionally (after a respiration trial) at 14.00.

The experimental period was divided in two subperiods. Feed residues were collected, weighed and sampled at the end of each subperiod.

As the composition of the faecal dry matter hardly varied in both subperiods the determinations of crude fat, crude fibre, ash, carbon and calorific value, from R 33 on, were carried out in a mixture of aliquote parts of the two air-dried samples of the subperiods.

II.3. THE HAY AND THE PREPARATION OF THE RATIONS

From four pastures of the laboratory experimental farm early and late cut hay were harvested. Each time two strips of one pasture furnished the early cut hay and one or two others the late cut hay. Table II 3 gives some data about the harvest of these hays.

TABLE II 3. Some data on the harvest of the hay

| Hay | Soil | Cutting date (A) | Weather between A and B | Put on frames (B) | Brought indoors | Heating in stack |
|---------------|-----------------|------------------|-------------------------|-------------------|-----------------|------------------|
| Early 1958 | } clay | 24 May | some rain | 29 May | 11 June | none |
| Late 1958 | | 20 June | several showers | 30 June | 9 July | hardly any |
| Early 1959 | } clay | 14 May | sunny, dry | 19 May | 1 June | none |
| Late 1959 | | 30 May | sunny, dry | 1 June | 12 June | none |
| Early 1960 I | } clay and peat | 10 May | sunny, dry | 12 May | 3 June | none |
| Late 1960 I | | 8/9 June | rainy | 18 June | 25 June | none |
| Early 1960 II | } clay and peat | 18 June | some drizzles | 23 June | 14 July | none |
| Late 1960 II | | 15 July | some showers | 21 July | 3 August | none |

Some weeks prior to an experiment the hay was chopped to pieces of a length of ± 5 cm and part of the chopped hay was sieved in two or three fractions. Each fraction was mixed thoroughly. The rations for the first two weeks of the preliminary period consisted of the chopped but not sieved hay. Those for the last week of the preliminary period and for the experimental period were weighed on one day; they were sampled by taking aliquote parts of the sieved fractions.

In addition to the hay the animals were given daily 30 g NaCl. In the first preliminary week of some experiments with animals not having been on pasture for several months vitamin A and D and copper and cobalt were given (in total per head: period R 29–32 1 125 000 IU vitamin A, 375 000 IU vitamin D₃, 14 g CuSO₄5aq. and 0.35 g CoSO₄7aq.; in period R 40 and R 41 the same was given but with double amount of vitamin A). The same quantities of copper and cobalt but no vitamins were given to the animals in the first preliminary week of the periods R 33–36, these animals having been on pasture for some months prior to R 33.

II.4. COLLECTION OF FAECES AND URINE

For collecting faeces and urine separately the urinals described by RICHTER and BECKER (1952) were used and fastened to the animals with a leather harness (VAN ES and VOGT, 1959). The faeces fell into big galvanised containers and the urine flowed through urinal and tubing into a 10-litre flask. Any urine which escaped collection in the urinal fell into the big container and was collected as free from faeces as possible. The amount of this spilled urine was seldom more than 5 % of the total quantity.

II.5. RESPIRATION TRIALS

Up to R 20 respiration trials were conducted on two subsequent days (48 hours) in the middle of each of the two subperiods. Afterwards, for obtaining a higher accuracy, these trials were performed for three 48-hour periods, one close by the start, one in the middle and one towards the end of the experimental period. All animals were accustomed to such experiments because in or before the preliminary period they stayed in the respiration chambers for several hours on various days. There was no significant difference between the time spent standing in the digestion stall and in the respiration chambers. Usually the animals were standing for slightly more than 12 hours a day.

For performing the respiration trials we had two identical open-circuit respiration chambers at our disposal. They were used almost in the same way as described earlier (VAN ES, 1961, p. 51). The temperature in the chambers was 15°C and the relative humidity close by 75 %.

Slight modifications with the aim to reduce the amount of work were introduced when the periods with four cows started (R 33).

II.6. WEIGHING, SAMPLING AND ANALYSIS OF FEED, FAECES AND URINE AND CALCULATION OF THE BALANCES

The methods of weighing, sampling and analysis have been described earlier (VAN ES, 1961, p.45). The determination of true protein was omitted. From period R 33 onward one additional sample of hay was taken and analysed for dry matter content in order to obtain a higher accuracy. Just as in the earlier periods R 1-14, in the present series the contents of dry matter and crude protein as determined in the wet faeces were used in the further calculations together with the contents of crude fat, crude fibre, ash, carbon and calorific value determined in the faeces dried at 60-70°C. In earlier experiments it had been shown that drying of faeces at 60-70°C which gave a loss of about 5% of the nitrogen did not result in a loss of the complete protein molecules but in a loss of more simple N-containing degradation products, the dry matter content practically remaining the same. Therefore, from R 19 onward corrections for loss of dry matter in drying were neglected.

The following constants were used in computing the N-content, the C-content and the calorific value of CO₂, CH₄, protein and body fat:

- 1 litre CO₂¹) contains 0.5362 g C,
- 1 litre CH₄¹) contains 0.5362 g C and yields 9.45 kcal in combustion,
- Protein contains 16% N, 52% C and yields 5.7 kcal/g in combustion,
- Body fat contains 76.73% C and yields 9.5 kcal/g in combustion.

The heat expenditure (H , kcal) was computed from O₂-consumption (O_2 , litres¹), CO₂-production (CO_2 , litres¹), urinary N (N , g) and CH₄-production (CH_4 , litres¹) with the formula (BROUWER, 1958):

$$H = 3.869 O_2 + 1.195 CO_2 - 0.227 \times 6.25 N - 0.516 CH_4.$$

The energy balance (G , kcal) was computed from metabolisable energy (M , kcal) and heat expenditure (H , kcal) and, moreover, from C-balance (C , g) and N-balance (N , g) with the formulae:

$$\begin{aligned} G_1 &= M - H, \\ G_2 &= 12.3785 C - 0.7368 \times 6.25 N, \\ G &= \frac{1}{2} (G_1 + G_2). \end{aligned}$$

The formalin used to preserve the samples of the faeces and the non-acidified samples of the urine was supposed to contain 40% formaldehyde, 16% C and 1.6 kcal/g.

The computation of the heat expenditure was also done with an electronic computer. The computation itself with this computer was very rapid. The

¹ 0°C, 760 mm Hg, dry.

collection of the data and the preparation of the punch tape used to transfer the data to the computer, however, took a considerable time. More time may be saved by this method when the data are punched by the respiration equipment itself.

II.7. ERRORS OF THE RESULTS DUE TO ANALYTICAL AND PHYSIOLOGICAL VARIATION

II.7.1. Introduction

Errors due to analytical variation are thought to include all errors made while weighing feed, feed residues, faeces and urine or measuring the total volume of outgoing air and while sampling and analysing the samples. Errors due to physiological variation include errors due to variation in daily production of faeces, urine, CO₂, CH₄ and heat or consumption of O₂ and errors due to daily changes in digestibility and in utilisation of the digested material.

Three methods were used to estimate these errors. An estimate (s) of the standard deviation of n independent variates $x_1, x_2, x_3, \dots, x_k, \dots, x_n$ with an average value \bar{x} was computed in the usual way:

$$s = \sqrt{\frac{\sum(x_k - \bar{x})^2}{n - 1}}$$

If only pairs of variates were available, i.e. $x_{11}, x_{12}; x_{21}, x_{22}; \dots; x_{k1}, x_{k2}; \dots; x_{n1}, x_{n2}$, the standard deviation of the average of one pair was estimated as

$$s = \sqrt{\frac{\sum(x_{k1} - x_{k2})^2}{4n}}$$

thus from the differences within the pairs. The third method was analysis of variance.

There were sufficient data for such computations. The hay for instance was sampled in duplicate and the resulting samples analysed separately. Moreover, the experimental periods usually were divided in two subperiods. Finally, each kind of hay was used in 4 or 6 experiments with different cows and the single experiments were part of reversal trials.

II.7.2. Feed intake

As already mentioned eight kinds of hay have been examined in 44 single experiments so that each kind of hay has been analysed several times. The analyses were performed in duplicate samples taken, independently of each other, during weighing the rations. In table II 4 are given the standard deviations of the averages calculated a) from within duplicate variation and b) calculated from between duplicate variation.

TABLE II.4. Analytical variation of the constituents of the hay (dry matter basis)

| | Standard deviations of averages of duplicates within hays | | | |
|-----------------|---|-----------|--|-----------|
| | Calculated from <i>within</i> duplicate variation | | Calculated from <i>between</i> duplicate variation | |
| | Average stand. dev. | Range | Average stand. dev. | Range |
| Dry matter | 0.18 % | 0.04-0.27 | 1.43 % | 0.45-2.30 |
| Crude protein | 0.06 % | 0.03-0.11 | 0.12 % | 0.06-0.17 |
| Crude fat | 0.16 % | 0.02-0.41 | 0.20 % | 0.07-0.44 |
| Crude fibre | 0.18 % | 0.05-0.27 | 0.61 % | 0.22-0.96 |
| N-free extract | 0.22 % | 0.07-0.43 | 0.82 % | 0.39-1.41 |
| Ash | 0.13 % | 0.04-0.26 | 0.21 % | 0.03-0.44 |
| Carbon | 0.10 % | 0.04-0.13 | 0.27 % | 0.07-0.46 |
| Calorific value | 8 cal/g | 6-12 | 18 cal/g | 7-31 |

The samples of each kind of hay, although coming from one big stock, differed slightly from one experiment to another since the standard error calculated from between duplicate variation is higher than the standard error calculated from variation within duplicates. It is clear that the dry matter content showed the highest between duplicate variation because the long intervals between the periods caused changes in moisture content of the hay. Between duplicate variation has also caused higher standard deviations of the components of the dry matter. This may easily be understood as the hay taken from the big stock for each single period (sometimes for each double period) was not a representative sample for the whole stock. Care was only taken that the hay came from the upper, the middle and the lower part of the stock. Slight changes in the analytical methods in the course of time may have contributed to this higher between duplicate variation. This might have been cleared up by comparing from time to time the results of current samples with those of a big sample analysed simultaneously.

The variation within the duplicates was slightly higher than that in earlier experiments (period R 1-14). However, with the intelligible exception of the dry matter content, even the *between* duplicate variation was not high.

II.7.3. Digestibility

Variation of the apparent digestion coefficients may be due to analytical, physiological, between period, between animal and between hay variation. In table II.5 the results are given of computations of the standard deviations of these coefficients a) within hays within single experiments computed from the results of the two subperiods of each single experiment and b) within hays between experiments. The former standard deviations are free from between

hay, between animal and almost free from between period variation; the latter are only free from between hay variation. The difference must therefore almost entirely be ascribed to between animal and between period variation.

Both sets of values and their ranges were only slightly different. It seems therefore correct to state that variation between animals and between periods was very small or absent.

TABLE II 5. Variation of digestibility

| | Standard deviation of apparent digestion coefficients | | | | | |
|-----------------|---|---------|----------|---------------------------------|-----------|-------------------|
| | Within experiments within hays | | | Between experiments within hays | | |
| | Average | | Range | Average | | Range |
| | R 1-14 | R 15-41 | R 15-41 | R 1-14 | R 15-41 | R 15-41 |
| Dry matter | 0.5 | 1.0 | 0.5-1.6 | 0.7 | 0.9 | 0.5-1.2 |
| Crude protein | 0.9 | 1.1 | 0.4-1.9 | 1.5 | 1.8 | 0.7-2.7 |
| Crude fibre | 0.7 | 1.0* | 0.4-1.2* | 1.0 | 1.0(1.0*) | 0.8-1.2(0.8-1.2*) |
| N-free extract | 0.4 | 1.1* | 0.4-1.6* | 0.7 | 1.5(1.1*) | 0.4-2.3(0.4-1.3*) |
| Organic matter | - | 1.0* | 0.4-1.5* | - | 0.9(0.9*) | 0.5-1.3(0.5-1.0*) |
| Calorific value | 0.5 | 1.2* | 0.5-1.7* | 0.7 | 1.0(0.9*) | 0.5-1.3(0.5-1.1*) |

* values of hays 1958 and 1959 only

The results of earlier experiments in period R 1-14 also suggested between animal variation in digestibility of only very small size. In these experiments a mixture of hay and concentrates was fed at the maintenance level and the variation within and between experiments was lower (table II 5) than in the present experiments. The higher variation in the present experiments may have been due to the fact that only hay was fed. It could be shown that in general in these hays the variation in digestibility of dry matter, organic matter, crude protein, N-free extractives and calorific value was higher for hays of lower digestibility. In experiments of BROUWER and DIJKSTRA (1938) in which the digestibility of nine kinds of hay was determined, for each hay with 2-3 bulls, we computed a within hay standard deviation of the digestion coefficient of the dry matter of 1.0 unit which differs little from the standard deviation in the present experiments.

On hay-rations long experimental periods appear to be necessary if highest accuracy is to be obtained, especially in case of hay with low digestibility.

II.7.4. Energy in urine and in methane; metabolisable energy

The energy in urine and in methane was computed for single experiments as a percentage of gross energy intake. The variation calculated from variance between single experiments within hays proved to be higher than that derived

from within single experiments as computed from differences between sub-periods, but in both cases it was small. In the earlier experiments (period R 1-14) it was only slightly lower (table II 6).

TABLE II 6. Variation of energy in faeces, urine and methane and of metabolisable energy

| | Standard deviation (% of energy intake) | | | | | |
|----------------------|---|---------|------------|---------------------------------|------------|-------------------|
| | Within experiments within hays | | | Between experiments within hays | | |
| | Average | | Range | Average | | Range |
| | R 1-14 | R 15-41 | R 15-41 | R 1-14 | R 15-41 | R 15-41 |
| Energy in faeces | 0.5 | 1.2* | 0.5 -1.7* | 0.7 | 0.97(0.9*) | 0.5-1.3(0.5-1.1*) |
| „ „ urine | 0.1 | 0.1* | 0.07-0.14* | 0.2 | 0.3 (0.3*) | 0.1-0.3(0.1-0.3*) |
| „ „ CH ₄ | 0.2 | 0.1* | 0.1 -0.2* | 0.2 | 0.3 (0.3*) | 0.2-0.5(0.2-0.5*) |
| Metabolisable energy | 0.4 | 1.3* | 0.7 -1.8* | 0.6 | 0.95(0.8*) | 0.4-1.3(0.4-1.3*) |

* values of hays 1958 and 1959 only

The variation of the metabolisable energy computed as a percentage of gross energy intake was close to the variation of the digestion coefficient of energy and, therefore, also higher than in the earlier experiments with the mixed rations. There was no evidence of between animal or between period variation of this percentage. The standard deviation again referring to one single experiment was 1% of the average intake of about 27000 kcal for the whole material, thus about 270 kcal.

Of course the gross energy intake of the early hay generally was smaller and that of the late hay was larger than this average of 27000 kcal, the mean intakes being 25000 and 29500 kcal respectively. Since, furthermore, higher digestibility tended to give a lower variation coefficient, we estimate the standard deviation of the metabolisable energy of early cut hay at ± 240 kcal and that of the late cut hay at ± 300 kcal.

II.7.5. Heat expenditure

The analytical accuracy of the respiration equipment was checked with several CO₂- and N₂-tests in the course of this series of trials. As mentioned in paragraph II.7.1 this analytical accuracy includes errors of measuring gas volumes etc. In each of 7 CO₂-tests between 98.6% and 99.9% of the introduced volume of CO₂ was recovered. In other tests pure N₂ was introduced into the chamber and supposed to be the residue of a quantity of fresh air from which the oxygen completely had been consumed. The outgoing air was measured and analysed and from these figures the apparent oxygen consumption was calculated and expressed as a percentage of the volume of the theoretically

consumed oxygen just mentioned. In each of 12 N₂-tests these percentages were lying between 99.1 % and 99.9 % and in one more test 101.8 % was found.

The daily variation in the heat production of the animals computed from gaseous exchange and urinary nitrogen, was calculated from differences in heat production on two successive respiration days, moreover on two respiration days with an interval of one week and finally on two respiration days with an interval of two weeks. This was done for the whole material as well as for separate hays and for separate animals. The tables II 7 and II 8 show the results expressed as standard deviations in kcal per trial of 24 hours.

The variation for the interval of one day (respiration trials on successive days) is clearly lower than the variation for the interval of one week. There is little difference between the variations for intervals of one and two weeks. Neither kind of hay nor individuality of animal influenced the variation.

In the earlier experiments of periods R 1-14 with only intervals of one day and one week materially the same was found as in the present experiments. Then, the coefficient of variation was slightly higher: 1.8 % and 3.0 % of the daily heat expenditure respectively as compared with 1.6 % and 2.4 % at present. As these values are much higher than the analytical variation found in testing the respiration equipment in the CO₂- and N₂-trials just mentioned, they contain physiological variation of considerable size.

From these figures we estimate the standard deviation of the heat production per single experiment with 4 and 6 respiration days at $2.5/\sqrt{4} = 1\frac{1}{4}$ % and $2.5/\sqrt{6} = 1.0$ % respectively of the average heat production, i.e. at 160 kcal and 130 kcal respectively. These values are considerably lower than the standard deviation of the metabolisable energy (240-300 kcal). However, these figures for the variation of heat expenditure apply only to within single experiments, those for metabolisable energy to within and between experiments.

II.7.6. *The nitrogen, carbon and energy balances*

The standard deviation of the N-balance within single experiments was computed from the differences between the balances of the subperiods. The result of R 19 with Alke fed late cut hay 1958 was excluded because this animal had an exceptionally high difference in digestibility between the subperiods. For the other experiments up to R 32 the standard deviation was $14/6.25 = 2.24$ g or 1.8 %, for those in period R 33-41 somewhat lower: $10/6.25 = 1.60$ g or 1.2 % of the average N-intake amounting to $800/6.25 = 128$ g N. In the earlier experiments of period R 1-14 the standard deviation was $7/6.25 = 1.12$ g.

All values were calculated from within single experiment (between subperiod) variance. From between experiment variance SCHIEMANN et al. (1961) have found a higher standard deviation of about 3 % of the intake with adult steers on a maintenance ration.

TABLE II 7. Variation in heat production within experiments within hays

| Interval used for computation | Standard deviation (kcal) of heat production per 24 hours | | | | | | | | | | | |
|-------------------------------|---|------|--|--------------------|------|--|--------------------|------|------------|------|-------------|------|
| | The same 6 animals | | | The same 4 animals | | | The same 6 animals | | | | | |
| | Hay 1958 | | | Hay 1959 | | | Hay 1960I | | Hay 1960II | | All hays | |
| | Early | Late | | Early | Late | | Early | Late | Early | Late | Early | Late |
| One day | 210 | 170 | | 300 | 220 | | 240 | 180 | 200 | 180 | 210 (1.6 %) | |
| One week | 210 | 440 | | 390 | 220 | | 340 | 350 | 240 | 260 | 310 (2.4 %) | |
| Two weeks | 570 | 170 | | 240 | 210 | | 350 | 370 | 280 | 260 | 320 (2.5 %) | |

TABLE II 8. Variation in heat production within experiments within animals

| Interval used for computation | Standard deviation (kcal) of heat production per 24 hours | | | | | | | | | | | | | |
|-------------------------------|---|--------|----------|--------|-----------------|-------|----------|--------|-----------------|----------|--------|-------|---------|---------|
| | The same 8 hays | | | | The same 4 hays | | | | The same 4 hays | | | | | |
| | Alke | | Zws. 2 | | Alke | | Zws. 2 | | Alke | | Zws. 2 | | Rooisje | |
| | R. Willy | Zws. 2 | R. Willy | Zws. 2 | Alke | Kee 2 | R. Willy | Zws. 2 | Alke | R. Willy | Zws. 2 | Kee 3 | Jansje | Rooisje |
| One day | 200 | 230 | 260 | 260 | 170 | 170 | 240 | 320 | 220 | 220 | 190 | 240 | 140 | 190 |
| One week | 190 | 420 | 280 | 280 | 190 | 410 | 390 | 270 | 190 | 440 | 280 | 250 | 360 | 180 |
| Two weeks | 260 | 450 | 180 | 180 | 210 | - | 480 | 110 | 280 | 420 | 220 | 260 | 400 | 280 |

For the other balances the calculation of the standard deviation per single experiment from subperiod differences was only possible in the experiments up to R 32 in which C-content and calorific value of feed, faeces, urine and also gaseous exchange were known for each subperiod. The standard deviations were for the C-balance: 31 g, for the energy balance computed as the difference between metabolisable energy and heat production: 305 kcal, and for the energy balance computed from C- and N-balance: 383 kcal. The average intakes of carbon and gross energy were 2800 g and 27000 kcal respectively.

In section II.7.4 and II.7.5 we have found that for a single experiment with six respiration days the standard deviation of the metabolisable energy was about 1 % of the gross energy intake and that of the heat production about 1 % of the daily heat production. From this a standard deviation of the energy balance of $\sqrt{270^2 + 140^2} = 310$ kcal may be computed which figure differs little from the figure 305 mentioned above. From this similarity it follows that on the same feed deviations of metabolisable energy and of heat expenditure are practically not correlated. In the earlier experiments in periods R 1-14 a standard deviation of 230 kcal had been calculated from gaseous exchange and urine-N (4 respiration days per single experiment). Then, the standard deviation of the energy balance computed from C- and N-balances (250 kcal) again was higher.

The correlation between the energy balances computed in the two ways mentioned above was very high ($r = 0.98$) which is usually the case (HOFFMANN, 1958; HOFFMANN et al., 1958; VAN ES, 1961). The average of the two balances, therefore, will have a standard deviation of about the same magnitude, i.e. about 350 kcal or 1.3 % of the gross energy intake. In computing these figures only within single experiment variation is taken into account.

The energy balances computed from C- and N-balances were usually higher than the balances computed as a difference between metabolisable energy and heat expenditure. In one experiment the difference amounted to +2.2 % of the gross energy intake, in eight between +1.5 and +1.1 %, in 27 between +1.0 and 0.0 % and in eight between 0.0 % and -1.0 %. Somewhat larger differences in the same direction (average 1.8 % and 1.6 %) were observed by NEHRING et al. (1961) and by HOFFMANN et al. (1962).

CHAPTER III

EXPERIMENTAL

Data referring to the individual cows on live weight, feed intake, digested feed constituents, digestion coefficients, urine, gaseous exchange, metabolisable energy and balances (N-, C- and energy balances) are given in table A at the end of this paper. The smaller tables are inserted in the text.

III.1. THE COMPOSITION OF THE HAYS

In table III.1 the average compositions of the eight kinds of early and late cut hay are given. On a dry matter basis the early cut hay had

| | 1958 | 1959 | 1960I | 1960II |
|---------------------------------|------|------|-------|--------|
| Higher percentage of protein | 8 % | 4 % | 8 % | 5 % |
| „ „ „ crude fat | 1.2% | 0.4% | 1.6% | 0.4% |
| „ „ „ ash | 2.9% | 1.4% | 0.6% | 0.5% |
| Lower percentage of crude fibre | 10 % | 3 % | 12 % | 4 % |
| „ „ „ N.f. extr. | 1.6% | 2.7% | -1.7% | 1.4% |

Therefore the aim to obtain early and late cut hays of clearly different composition was achieved except for the hay of 1959. In that year a severe drought stopped the growth of the sward after the early hay had been cut.

The content of protein on a dry matter basis was only in one case (late hay 1958) lower than the average content (10.35 %) of hay in the Netherlands given by BROUWER and DIJKSTRA (1938), in all other cases higher or even much higher; the reverse was true with the crude fibre content which the just mentioned investigators found to be 34.73 %.

The differences in calorific value between the early and the late hays were small if computed on a dry matter basis, but greater, from +2 up to +4 %, if computed on an organic matter basis. This fact may be explained by the higher content of crude protein and of crude fat of the early cut hay only partly compensated by the higher lignine content of the late cut hay. All three constituents have a high calorific value compared with carbohydrates.

III.2. FEED RESIDUES

Feed residues were at most 1.1 % when early cut hay was fed. Residues of some size (9, 19, 11, 19, 3 and less than 1 % respectively) were left by the six animals fed with late cut hay 1959. All other residues were at most 1 % of the daily ration with the exception of those of two animals fed late hay 1960II (4 and 8 % respectively).

TABLE III.1. Composition of the hays

| Hays | Dry matter (%) | On a dry matter basis | | | | | | | | | | Calorific value of organic matter (cal/g) |
|----------------------------|----------------|-----------------------|-------------------|---------------|-----------------|------------------|---------|-------|-------------------------|------|--|---|
| | | Organic matter (%) | Crude protein (%) | Crude fat (%) | Crude fibre (%) | N-free extr. (%) | Ash (%) | C (%) | Calorific value (cal/g) | | | |
| Early '58 | 86.30 | 88.07 | 16.56 | 2.97 | 27.92 | 40.62 | 11.93 | 43.64 | 4280 | 4860 | | |
| Late '58 | 86.97 | 90.93 | 8.51 | 1.81 | 38.34 | 42.26 | 9.07 | 44.62 | 4298 | 4727 | | |
| Early '59 | 88.32 | 90.76 | 16.01 | 3.30 | 24.49 | 46.95 | 9.24 | 45.04 | 4425 | 4875 | | |
| Late '59 | 89.60 | 92.14 | 11.81 | 2.92 | 27.78 | 49.62 | 7.86 | 45.10 | 4413 | 4789 | | |
| Early '60I | 84.50 | 91.03 | 19.89 | 3.71 | 21.54 | 45.90 | 8.97 | 45.55 | 4523 | 4969 | | |
| Late '60I | 85.44 | 91.67 | 11.60 | 2.10 | 33.79 | 44.18 | 8.33 | 45.12 | 4389 | 4788 | | |
| Early '60II | 86.72 | 89.89 | 18.72 | 2.45 | 28.92 | 39.80 | 10.11 | 44.88 | 4448 | 4948 | | |
| Late '60II | 86.12 | 90.35 | 13.77 | 2.01 | 33.36 | 41.21 | 9.65 | 44.73 | 4375 | 4842 | | |
| SD ¹⁾ | 0.18 | 0.13 | 0.06 | 0.16 | 0.18 | 0.22 | 0.13 | 0.10 | 8 | | | |

¹⁾ S D = average standard deviation within experiments within hays; the standard deviation of the average is not given; only 3 figures were available for each hay 1958 and 4 for the other hays

TABLE III.2. Digestibility, energy losses in faeces, urine and methane and metabolisable energy %

| Hays | Digestibility percentage | | | | | | | Energy loss as a % of gross energy intake in | | | | Metab. energy as a % of dig. energy |
|----------------------------|--------------------------|----------------|---------------|-------------|--------------|--------------|--------|--|---------|---------------|------|-------------------------------------|
| | Dry matter | Organic matter | Crude protein | Crude fibre | N-free extr. | Gross energy | Faeces | Urine | Methane | Metab. energy | | |
| | Early '58 | 71.2 | 74.7 | 68.3 | 81.5 | 74.2 | 70.2 | 29.8 | 6.3 | 8.5 | 55.4 | |
| Late '58 | 58.4 | 59.9 | 46.9 | 67.3 | 56.8 | 55.0 | 45.0 | 3.4 | 7.9 | 43.8 | 79.6 | |
| Early '59 | 73.2 | 76.3 | 68.0 | 80.2 | 78.3 | 71.8 | 28.2 | 6.6 | 8.4 | 56.8 | 79.1 | |
| Late '59 | 70.2 | 72.4 | 62.8 | 72.6 | 75.6 | 68.0 | 32.0 | 5.5 | 8.3 | 54.2 | 79.7 | |
| Early '60I | 76.7 | 79.1 | 73.6 | 83.0 | 81.4 | 74.7 | 25.3 | 7.2 | 8.3 | 59.1 | 79.1 | |
| Late '60I | 59.0 | 60.8 | 57.6 | 64.9 | 59.5 | 56.3 | 43.7 | 4.2 | 7.4 | 44.8 | 79.6 | |
| Early '60II | 63.6 | 65.6 | 66.4 | 72.9 | 61.7 | 60.7 | 39.3 | 6.2 | 7.5 | 47.0 | 77.4 | |
| Late '60II | 53.1 | 54.5 | 55.4 | 60.6 | 50.5 | 50.0 | 50.0 | 4.1 | 6.7 | 39.2 | 78.4 | |
| SD ¹⁾ | 0.9 | 0.9 | 1.8 | 1.0 | 1.5 | 0.97 | 0.97 | 0.3 | 0.3 | 0.95 | | |

¹⁾ SD = average standard deviation between experiments within hays; the standard deviation of the average is not given; only 4 experiments have been done with hays 1959 and 6 with the other hays

III.3. DIGESTIBILITY

In table III2 a survey is given of the average digestibility of the hay components. As was to be expected from the composition there was a considerable difference between the early and late cut hays with the exception of the hays of 1959.

The range from lowest to highest digestibility was for dry matter 53-77, for organic matter 54-79, for energy 50-75, for crude protein 47-74, for crude fibre 61-83 and for N-free extractives 50-81.

We computed for all the hays the average contents of digestible crude protein and of starch equivalent in the dry matter, the latter from the contents of the digestible organic constituents and of crude fibre with the method of KELLNER (table III3). If ranked after increasing values the sequence of the digestible crude protein closely follows that of the crude protein content and the sequence of the starch equivalent follows that of the digestibilities. The range of the former is 4-15% and of the latter 29-58%. The agreement with the figures computed by regression with the aid of the revised formulae of BROUWER and DIJKSTRA used in the Netherlands¹ (table III3) is satisfactory taking into consideration the errors adherent to such a method.

TABLE III3. Content of digestible crude protein and of computed starch equivalent of the dry matter of the hay

| | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|---|----------|------|----------|------|-----------|------|------------|------|
| | Early | Late | Early | Late | Early | Late | Early | Late |
| % digestible crude protein in dry matter | 11.3 | 4.0 | 10.9 | 7.4 | 14.6 | 6.7 | 12.4 | 7.6 |
| The same, computed from regression on crude protein | 10.3 | 3.7 | 9.8 | 6.4 | 12.9 | 6.2 | 11.9 | 8.0 |
| Computed content of starch equivalent | 49 | 32 | 55 | 50 | 58 | 36 | 41 | 29 |
| The same, computed from regression on crude fibre . | 42 | 27 | 50 | 46 | 56 | 36 | 42 | 35 |

According to the figures of digestible crude protein and starch equivalent given by KELLNER-BECKER (1959) the early hays of 1958, 1959 and 1960I, and the late hay 1959 were of best quality or even better. The early hay 1960II was of very good, the late hay 1960I was of good and the late hays 1958 and 1960II were of moderate quality.

¹ Manual for the calculation of the nutritive value of roughages, Bedrijfslaboratorium voor Grond- en Gewasonderzoek Mariëndaal, Oosterbeek, the Netherlands (1958).

It should be kept in mind that year of harvest, pasture and weather conditions varied for the different sets of early and late cut hay. Stress, therefore, should be laid more upon the differences in properties within each set than between sets. Except for the hay 1959 the differences within the sets are considerable.

III.4. METABOLISABLE ENERGY; ENERGY LOSSES WITH URINE AND METHANE

Table III 2 gives a survey of the average losses of energy with urine and methane per 100 kcal of gross energy intake. All losses are higher for the early cut hay, especially those with the urine. Therefore, per 100 g of dry matter intake the difference in content of metabolisable energy between early and late cut hay is smaller than the difference in digestible energy. Per 100 g of digestible dry matter, however, the difference between digestible and metabolisable energy in early cut hay is nearly equal to that in late cut hay because the greater difference per 100 g of dry matter in the early cut hay coincides with a higher percentage of digestible dry matter. This is the reason why the ratios between metabolisable and digestible energy are almost the same for all hays (table III 2, last column).

Correlation coefficients were computed between the energy in the urine (% of gross energy) and 1) the crude protein content (%) of the dietary dry matter, 2) the crude fibre content (%) of the dietary dry matter, and 3) the digestibility (%) of the dietary energy. All these correlations were high and significant: $r = +0.88$, $r = -0.96$ and $r = +0.89$ respectively. The strong negative correlation with crude fibre may not be explained by correlation between content of crude protein and crude fibre ($r = -0.81$) only; evidently the negative correlation between crude fibre and digestibility is also important.

The two regression equations referring to energy in urine (Y) as a percentage of gross energy intake on crude fibre percentage of the dry matter (X_1) and on the digestion coefficient of gross energy (X_2) were as follows:

$$Y = (-0.24 \pm 0.03)X_1 + 12.65; \quad s_{YX_1} = 0.4,$$

$$Y = (+0.13 \pm 0.03)X_2 + 3.13; \quad s_{YX_2} = 0.7.$$

The loss of energy in methane was computed per 100 kcal of gross energy and of digestible energy, furthermore per 100 g of dry matter, of organic matter and of some components of the dry matter, and finally per 100 g of digestible dry matter and of some of its components.

The averages of the first 6 lines are close to those given by WÖHLBIER and SCHNEIDER (1961) and to those of KELLNER and FINGERLING quoted by them. In all cases their coefficients of variation were somewhat lower than ours; both their and our coefficients referring to the relation with crude fibre are high.

| | Methane | | | Coefficient of variation (%) of methane |
|--------------------------------|---------|-------|-------|---|
| | kcal | g | litre | |
| Per 100 kcal gross energy | 7.88 | 0.60 | 0.83 | 7.6 |
| Per 100 g dry matter | 34.60 | 2.62 | 3.66 | 8.1 |
| „ 100 „ organic matter | 38.20 | 2.89 | 4.04 | 8.4 |
| „ 100 „ crude fibre | 122.00 | 9.24 | 12.91 | 25.5 |
| „ 100 „ NFE | 79.20 | 6.00 | 8.38 | 7.7 |
| „ 100 „ crude fibre and NFE | 47.50 | 3.60 | 5.03 | 12.6 |
| „ 100 „ digestible dry matter | 53.00 | 4.02 | 5.61 | 5.7 |
| „ 100 „ digest. organ. matter | 56.60 | 4.29 | 5.99 | 5.8 |
| „ 100 „ „ crude fibre | 165.40 | 12.53 | 17.50 | 15.7 |
| „ 100 „ „ NFE | 120.20 | 9.11 | 12.72 | 15.5 |
| „ 100 „ „ (cr. fibre + NFE) | 68.30 | 5.17 | 7.23 | 4.4 |
| Per 100 kcal digestible energy | 12.60 | 0.95 | 1.33 | 7.9 |

In the above table the lowest coefficient of variation (4.4 %) was found by relating methane to digestible carbohydrates (digestible crude fibre + digestible NFE): 5.17 g of methane per 100 g of digestible carbohydrates. In his textbook KELLNER (1919) gives a figure of 4.29 g/100 g of digestible carbohydrates and maintains that much higher figures are obtained by feeding wheat straw only. KLEIBER et al. (1945) found a value of 4.4 g/100 g of digestible carbohydrate on rations of Sudan hay. ARMSBY and FRIES (1915) came to 4.8 g/100 g for roughages and 4.7 g/100 g for mixed rations. In KLEIBER's experiments as well as in ours and those of WÖHLBIER et al. the methane energy was approximately 8 % of the gross energy.

In agreement with BLAXTER's results (1961) we also found slightly lower methane-energy losses per 100 kcal of gross energy for feeds of low digestibility. For rations consisting of roughage only he computed the following regression equation referring to methane energy as a percentage (Y_1) of gross energy intake on digestibility (X_1) of the gross energy:

$$Y_1 = 0.059X_1 + 4.28.$$

In our material we found almost the same equation:

$$Y_1 = (0.062 \pm 0.013)X_1 + 3.98; \quad s_{Y_1X_1} = 0.32.$$

We also computed the regression of methane energy (Y_2 , kcal) per 100 g of ingested dry matter on digestible dry matter (X_2 , g) per g of ingested dry matter, because the ratio between these items has a low coefficient of variation as was explained above:

$$Y_2 = (32.3 \pm 4.4)X_2 + 13.4; \quad s_{Y_2X_2} = 1.0.$$

III.5. THE N-, C- AND ENERGY BALANCES

In many experiments the N-balances were negative. The average crude protein and energy balances were as follows.

| | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|---|----------|--------|----------|-------|-----------|--------|------------|--------|
| | early | late | early | late | early | late | early | late |
| 6.25 × N-balance ± st.dev. | -6±19 | -40±44 | 0±18 | -1±34 | +16±28 | -44±31 | -19±27 | -21±39 |
| g digestible crude pro- tein intake per 100 kg bodyweight | 128 | 53 | 109 | 79 | 131 | 73 | 127 | 90 |
| kcal energy balance × (500/W) ^{3/4} | +1077 | +223 | +1068 | +1118 | +768 | -579 | -480 | -1352 |

Only for the late hay 1958 the intake of digestible crude protein was below the feeding standard of 70 g per 100 kg bodyweight. For all early hays the intake was considerably above this standard. The negative N-balances of the late hays 1960I and 1960II and of the early hay 1960II may have been caused by insufficient supply of energy since in nearly all experiments with these hays the energy balances were negative.

The C-balance (g) mainly determined the energy balance (kcal) since the latter is equal to 12.3785 times the C-balance minus 0.7368×6.25 times the N-balance (g) and because the N-balances were small as compared with the C-balances. Few energy balances were above +2000 kcal or below -2000 kcal. In 25 of the 44 experiments they were between +1000 and -1000 kcal. If estimates of the digestibility of the hays and of the animal's maintenance requirement of metabolisable energy had been available beforehand, it would have been possible to feed such quantities of hay that the energy balances would have been nearer to zero. For this reason preliminary digestion trials with sheep and computation of the result of the first balance experiment with a new cow as soon as this experiment is finished seem to be very useful.

CHAPTER IV

REGRESSION ANALYSIS

IV.1. METHODS

In order to get full information on the requirement of the cows for maintenance, and on the maintenance values of hays *regression formulae* were calculated demonstrating the regression between metabolisable energy, energy balance, and crude fibre consumed.

As mentioned before the experiments were performed in 9 double periods, every double period consisting of two single periods succeeding each other closely. In every double period two or four parallel double experiments covering the whole double period were run simultaneously, with one cow in each of the double experiments. In total there were 22 double experiments (with 44 single experiments); the number of cows was nine.

At the beginning of every double period two double experiments with one cow each were started at the same time, one with early cut hay and the other with late cut hay from the same field. After the first single period the hays were reversed and a transition period of 18–20 days was inserted, after which the second period followed. Thus, the experiments were arranged according to the reversal method in blocks of four single experiments. In some double periods there were run two blocks of four single experiments (two reversal trials).

Using KLEIBER's formula (1932, 1938, 1961) the observational data (energy balance, crude fibre consumed and metabolisable energy) were reduced to 500 kg live weight by multiplying by $(500/W)^{3/4}$ in which W = live weight (kg). Negative balances had been multiplied by 0.83 because for prevention of energy loss, less metabolisable energy is required than for body gain (MØLLGAARD, 1929).

It is true that in the opinion of some authors the exponent $3/4$ in the just mentioned formula should be higher or lower. However, VAN ES (1961) showed clearly from statistical analysis of respiration trials in the literature that variation of the exponent between 0.7 and 1.0 does not materially affect the regression coefficients. It is however also true that in our experiments the maintenance requirement would have been found somewhat lower if, for instance, the exponent 1.0 had been used instead of $3/4$. With the average live weight of 569 kg the ratio would have been

$$\frac{500}{569} : \left(\frac{500}{569}\right)^{3/4} = 0.968,$$

thus, a difference of about 3 %; this is also found in chapter V.

Another question is whether the ratio 0.83 (MØLLGAARD, 1929) between requirement for gain and requirement for prevention of negative gain (= requirement for maintenance) is correct.

VAN ES, in his statistical analysis, calculated the following partial regression coefficients between gain (G , kcal), metabolisable energy (M , kcal) and live weight (W , kg).

With negative balances: $b_{MG.W} = 1.4$, $1/b_{GM.W} = 2.0$,

With positive balances: $b_{MG.W} = 1.6$, $1/b_{GM.W} \leq 2.8$.

The ratios are therefore 0.88 and ≥ 0.71 with average ≥ 0.80 , a figure differing only slightly from MØLLGAARD's figure 0.83.

Some formulae of BLAXTER (1961, 1962) give a lower result for crude fibre rich feed such as hay. He maintains that the expenditure of energy in maintenance and muscular work takes place with a relative constant efficiency, the requirement of metabolisable energy per kcal of net energy for maintenance being 1.313, corresponding with an efficiency of 76 %. The efficiency of fat synthesis, however, was found by him to be related to the percentage of the gross energy of the ration which is metabolisable. In our hays this percentage was 49.05 % on an average. From BLAXTER's formula follows an efficiency of only 38.1 %, corresponding with a requirement of 2.62 kcal per kcal of gain. The ratio between the two requirements, therefore, would be only $1.313/2.62 = 0.50$. However, a ratio 0.83 fits better in with our data as will be shown in section IV.4.

The data were used for calculating the constants in the following regression formula:

$$M_{m500} + aG_{500} + b(F_{500} - \bar{F}_{500}) = M_{500}, \quad (\text{IV } 1)$$

where the suffix 500 refers to the live weight = 500 kg; furthermore

G_{500} = energy balance (kcal) (negative balances multiplied by 0.83),

F_{500} = crude fibre consumed (g), with average \bar{F}_{500} ; in our data $\bar{F}_{500} = 1717$ g,

M_{500} = metabolisable energy (kcal) ingested,

$M_{m,500}$ = maintenance requirement of metabolisable energy (kcal) with zero balance and crude fibre intake = $\bar{F}_{500} = 1717$ g,

a and b are constants to be computed.

In many computations we used the same formula (IV 1) but with a somewhat differently defined G , allowing for the fact that according to KELLNER energy in body fat is more efficiently produced from metabolisable energy in crude fibre poor rations than from energy in crude fibre rich rations. Therefore, the energy balance G_w actually found in a cow with live weight W is multiplied

by a factor c_s/\bar{c}_s so that we get $(c_s/\bar{c}_s)G_w$, in which \bar{c}_s is the mean value of c_s , and

$$c_s = \frac{M_w}{2365S_w} = \frac{M_{500}}{2365S_{500}} \quad (\text{IV } 2)$$

W and 500 stand for the live weight (kg),

M for the metabolisable energy (kcal) of the ration as found in the experiment, and

S for kilo-starch-units of the ration as calculated by KELLNER's method;

finally $S_{500} = (500/W)^{3/4} S_w$ and $M_{500} = (500/W)^{3/4} M_w$.

As \bar{c}_s represents the average of c_s , it is clear that the average value of $(c_s/\bar{c}_s)G$ is of the same order of magnitude as the average of G . The advantage of the outlined procedure is therefore that the coefficients a of G and of $(c_s/\bar{c}_s)G$ need no transformation but can directly be compared with each other.

In the literature there has been a great deal of discussion about the question whether, and if so in which way, a correction for standing and lying should be applied to G_w . Therefore, one set of calculations was performed with correction of the energy balance to 12 hr standing and lying and another without such a correction. The correction was based on the assumption that the heat expenditure in standing is 10 % higher than in lying. The correction formula was as follows:

$$u = \frac{U_s - 12}{U_s + 228} \times H. \quad (\text{IV } 3)$$

U_s = hours standing; fractions of hours in decimals,

H = heat expenditure (kcal),

u = correction (kcal) to be added to the energy balance.

We preferred correction to 12 hr standing because most cows spent about half a day in standing. Correction to 24 hr rest would therefore imply an extrapolation with increased risk of errors.

First of all, by the method of least squares, one formula (IV 1) with only three unknowns, $M_{m,500}$, a and b , was adjusted to the data of all 44 single experiments. This procedure was varied in four ways:

Method 1. Balance not corrected to 12 hr standing and not corrected with the use of c_s/\bar{c}_s .

Method 2. Balance corrected to 12 hr standing but not corrected with the use of c_s/\bar{c}_s .

Method 3. Balance not corrected to 12 hr standing but corrected with the use of c_s/\bar{c}_s .

Method 4. Balance corrected to 12 hr standing and also corrected with the use of c_s/\bar{c}_s .

TABLE IV 1. Constants of regression formulae

| Table IV 1a | | | | |
|---------------|----------------|----------------|--------------------------|-------|
| Method | $a = b_{MG.F}$ | $b = b_{MF.G}$ | $\bar{M}_{m,500}$ (kcal) | s^1 |
| 1. | +1.1206±0.120 | +0.6625±0.253 | 11907 | 799 |
| 2. | +1.1535±0.117 | +0.6679±0.243 | 11934 | 767 |
| 3. | +1.1118±0.118 | +0.5870±0.249 | 11970 | 793 |
| 4. | +1.1474±0.114 | +0.5996±0.238 | 11999 | 757 |
| Table IV 1b | | | | |
| 1. | +1.4612±0.150 | +0.8307±0.239 | 12009 | 665 |
| 2. | +1.5074±0.140 | +0.8400±0.222 | 12045 | 618 |
| 3. | +1.4364±0.141 | +0.7152±0.226 | 12102 | 644 |
| 4. | +1.4814±0.132 | +0.7331±0.209 | 12139 | 596 |
| Table IV 1c | | | | |
| 1. | +1.1831±0.170 | +0.3828±0.227 | 11893 | 427 |
| 2. | +1.2011±0.165 | +0.4144±0.223 | 11925 | 414 |
| 3. | +1.1309±0.155 | +0.2758±0.209 | 11966 | 413 |
| 4. | +1.1482±0.152 | +0.3145±0.207 | 11998 | 403 |
| Table IV 1d | | | | |
| 1. | +1.4335±0.150 | +0.6015±0.191 | 11837 | 338 |
| 2. | +1.4348±0.143 | +0.6218±0.183 | 11880 | 323 |
| 3. | +1.3085±0.146 | +0.4165±0.187 | 11937 | 357 |
| 4. | +1.3166±0.141 | +0.4519±0.183 | 11976 | 344 |
| Average IV 1d | +1.3734±0.145 | +0.5229±0.186 | 11908 | 340 |
| Table IV 1e | | | | |
| 1. | +1.5163±0.152 | +0.6790±0.185 | 11819 | 313 |
| 2. | +1.5170±0.143 | +0.6999±0.177 | 11864 | 296 |
| 3. | +1.3568±0.152 | +0.4593±0.188 | 11929 | 345 |
| 4. | +1.3664±0.146 | +0.4970±0.183 | 11969 | 331 |
| Average IV 1e | +1.4391±0.148 | +0.5838±0.183 | 11895 | 321 |

¹) Residual standard deviation.

The results are given in table IV 1a. Practically, they are the same for all four methods of calculation. $M_{m,500}$ varies only from 11.91 to 12.00 megacalories. The residual standard deviation s of one M_{500} -value above or below the regression plane is however rather large (757-799); the smallest value was obtained with the method 4. For the standard deviation of $M_{m,500}$ itself, not recorded in the table, we found 123, 118, 121 and 115 respectively; a proves to be highly significant and also b seems to be significant. All these results are however questionable, because the data on which the calculations are based, are lacking independence. For instance, between animal variations have not been taken into account.

Therefore, in the next calculation the between animal variation was allowed for by using 9 formulae (IV1) with 9 values $M_{m,500}$, one for each single cow:

$$\begin{aligned}M_{m,500,1} + aG_{500,1} + b(F_{500,1} - 1717) &= M_{500,1}, \\M_{m,500,2} + aG_{500,2} + b(F_{500,2} - 1717) &= M_{500,2}, \\&\dots \dots \dots \\M_{m,500,9} + aG_{500,9} + b(F_{500,9} - 1717) &= M_{500,9}.\end{aligned}$$

For all cows, a and b were considered to be equal, because they only occur in terms used by us for minor corrections. Thus, the total number of unknowns in these formulae amounted to $9 + 2 = 11$.

These formulae can be combined into one single formula:

$$\begin{aligned}&\alpha_1 M_{m,500,1} + \alpha_2 M_{m,500,2} + \dots + \alpha_9 M_{m,500,9} + \\+ a \{ &\alpha_1 G_{500,1} + \alpha_2 G_{500,2} + \dots + \alpha_9 G_{500,9} \} + \\+ b \{ &\alpha_1 (F_{500,1} - 1717) + \alpha_2 (F_{500,2} - 1717) + \dots + \alpha_9 (F_{500,9} - 1717) \} = \\= &\alpha_1 M_{500,1} + \alpha_2 M_{500,2} + \dots + \alpha_9 M_{500,9}.\end{aligned}$$

For cow k all α 's are zero, except α_k which is interpreted as 1.

The unknowns are $M_{m,500,1}$, $M_{m,500,2}$, ..., $M_{m,500,9}$ and furthermore a and b . They were computed with the method of least squares by adjustment to the data of all 44 single experiments, again with the four methods (1, 2, 3 and 4) mentioned before. The values for a , b and s , and the average ones for $M_{m,500}$, i.e. $\bar{M}_{m,500}$, are collected in table IV 1b. These results might also have been obtained with analysis of covariance.

Again, the results of the four methods of computation are quite similar and not very different from those in table IV 1a. a as well as b are significant. The average values $\bar{M}_{m,500}$ are 100 to 140 kcals higher than those in table IV 1a; the residual standard deviation s is somewhat reduced.

TABLE IV2. Individual maintenance requirements of metabolisable energy (kcal, 500 kg live weight) (period variation not eliminated; ordinary regression)

| Cow | No correction to 12 hr stand. without c_s | With correction to 12 hr stand. without c_s | No correction to 12 hr stand. with c_s | With correction to 12 hr stand. with c_s |
|--------------------------|---|---|--|--|
| 1. Zwartschoft 1 | 12157 | 12209 | 12243 | 12294 |
| 2. Zwartkop 2 | 12391 | 12486 | 12541 | 12619 |
| 3. Kee 2 | 11641 | 11524 | 11828 | 11717 |
| 4. Alke | 11429 | 11388 | 11568 | 11529 |
| 5. R W 12 | 11522 | 11478 | 11559 | 11523 |
| 6. Zwartschoft 2 | 11390 | 11609 | 11419 | 11637 |
| 7. Kee 3 | 11484 | 11583 | 11556 | 11660 |
| 8. Jansje | 12936 | 12962 | 13005 | 13032 |
| 9. Roosje | 13127 | 13165 | 13197 | 13241 |
| Average | 12009 | 12045 | 12102 | 12139 |

The individual maintenance requirements are given in table IV 2. They seem to show appreciable between animal variation, the figures for the upper two cows (1 and 2) and lower two animals (8 and 9) being markedly higher than those of the other animals. It is however worthy of note that cow 1 and 2 were tested in the earliest experiments of the series and 8 and 9 in the last ones, which suggests that a period variation might intervene.

We therefore had to examine whether between double period variations might obscure the real state of affairs. With this in mind, in order to eliminate between double period variation, a new computation with the method of least squares was set up in which the number of $M_{m,500}$'s was extended to 22, namely one for each cow in each double experiment. Again a and b were supposed to be the same for all the cows. Thus, there were $22 + 2 = 24$ unknowns which had to be calculated in the adjustment to the 44 figures for metabolisable energy as experimentally found in the 44 single experiments. Virtually, this way of computation fitted more closely to the design of the experiments because in every double experiment early and late cut hay from the same field were compared using the same cow. It should however be kept in mind that the range of the figures referring to crude fibre consumption is reduced which might result in lower regression coefficients. The regression formula used was as follows:

$$M_{m,500,ij} + aG_{500,ijt} + b(F_{500,ijt} - 1717) = M_{500,ijt}, \quad (\text{IV4})$$

where the suffix i stands for the double period, j for the number of the cow used in this double period, and t for the first and second single period of the double period i ($t = 1$ or 2). As already mentioned, the number of double periods amounted to 9 ($i = 1$ to 9); the total number of cows was also 9, but only two or four cows were used in one double period; therefore $j = 1, 2, 3$ or 4 .

Again, a , b , $\bar{M}_{m,500}$ and s were computed with the methods mentioned before. The results are collected in table IV 1c.

There are some remarkable differences between these results and those in table IV 1b. All the values of a are lower. Nevertheless a remains highly significant. Still more striking are the values of b as compared with the values in table IV 1b. They are reduced to about one half and hardly can be considered as significant. Presumably these differences are mainly caused by the elimination of the between double period variation.

So far, only the period variations between the different double periods have been eliminated, those *within* the double periods (between single periods) have been neglected. It will however be shown that the within variations, that means the differences between the two adjacent single periods of a double period may be large enough to affect markedly the results. In order to allow for these

within double period variations, calculations were made with a formula closely related to (IV 4).

In this new formula the maintenance requirement of cow j in the first part of the double period i is designated by $M_{m,500,ij} + v_i$, and in the second part by $M_{m,500,ij} - v_i$. The difference is therefore $2v_i$, a value considered as identical for all the cows taking part in the experiments conducted in double period i , but different for the different double periods.

The general formula is thus:

$$M_{m,500,ij} + (-1)^{t+1} v_i + aG_{500,ijt} + b(F_{500,ijt} - 1717) = M_{500,ijt}, \quad (\text{IV } 5)$$

in which t is interpreted as 1 and 2 for the first and second part of the double period under consideration. Again M = metabolisable energy, G = gain, F = crude fibre. The formula differs from (IV 4) only with respect to v_i , representing the within double period variation.

The two unknowns a and b are again the same for all the cows. Furthermore, there are again 22 $M_{m,500,ij}$'s and moreover 9 v 's, thus 33 unknowns in total. This formula was adjusted to the results of all 44 single experiments. There remained therefore only 11 degrees of freedom.

The number of normal equations was 33. After the elimination of the $M_{m,500,ij}$'s and v 's there remain two equations:

$$\left. \begin{aligned} Pa + Qb &= S, \\ Qa + Rb &= T, \end{aligned} \right\} \quad (\text{IV } 6)$$

from which a and b can be solved, P , Q , R , S and T being known.

It can be shown that these equations also can be written as follows:

$$\left. \begin{aligned} [G'G']a + [G'F']b &= [G'M'], \\ [G'F']a + [F'F']b &= [F'M'], \end{aligned} \right\} \quad (\text{IV } 7)$$

in which $[G'G']$ etc. have the following meaning.

Just as in formula (IV 4) and (IV 5) the suffix i will stand for the number of the double period ($i = 1, 2, \dots, 9$), j for one of the cows used in this double period ($j = 1, 2, 3$ or 4) and t for the first and second part of the double period ($t = 1$ or 2).

If only two cows j and k are used in a double period i the differences $G_{500,ij1} - G_{500,ij2} = G'_{ij}$ and $G_{500,ik1} - G_{500,ik2} = G'_{ik}$ are calculated. Then we compute a value G'_{ij} as follows:

$$G'_{ij} - 1/2\{G'_{ij} + G'_{ik}\} = 1/2\{G'_{ij} - G'_{ik}\} = G'_{ij},$$

and similarly for G'_{ik} .

For a double period i with four cows (j, k, l, m) we get

$$G'_{ij} - 1/4\{G'_{ij} + G'_{ik} + G'_{il} + G'_{im}\} = G'_{ij},$$

and similarly for G'_{ik} , G'_{il} and G'_{im} .

In putting $F_{500,ij1} - F_{500,ij2} = F'_{ij}$ and $M_{500,ij1} - M_{500,ij2} = M'_{ij}$, similar formulae can be obtained for F'_{ij} and M'_{ij} . Calculation of the squares and double products of G'_{ij} , F'_{ij} , and M'_{ij} , and addition over all the 22 double experiments yields the formulae (IV 7), from which a and b can be solved. It can be shown that $[G'G'] = 2P$, $[G'F'] = 2Q$, $[G'M'] = 2S$ etc.

As a matter of fact the equations (IV 6) and (IV 7) would also have been obtained in studying the covariance of G'_{ij} , F'_{ij} and M'_{ij} . These equations, therefore, also can be calculated with the aid of the differences G'_{ij} , F'_{ij} and M'_{ij} instead of by elimination of the M_m 's and v 's from the 33 normal equations. In this way a considerable amount of labour is saved.

The results obtained with (IV 5) are given in table IV 1d. The s -values have indeed improved a fairly good deal. The average values $\bar{M}_{m,500}$ are about the same (again a little below 12000 kcal). The values of a and b are a little higher than those in table IV 1c and their standard error is somewhat reduced so that a as well as b are significant. Since the formulae (IV 5) and (IV 7) gave the most favourable results the calculations with these and allied formulae were continued and most of the discussions in the following pages will be based on the results obtained in this way.

The figures for $a = b_{MG.F}$ in table IV 1d show that the requirement of metabolisable energy for body gain is about 1.37 kcal for 1 kcal of gain. The reciprocal is 0.73 so that one could think that this figure would represent the efficiency of metabolisable energy for gain. As a matter of fact the regression coefficients have a tendency to undervalue the ideal relationship between the variables and, consequently, the reciprocals tend to be too high. These reciprocals ($1/b_{MG.F}$) are given in the second column of table IV 3a. They are indeed always higher than the efficiencies calculated as the partial regression coefficients G on M , i.e. $b_{GM.F}$ in the third column.

TABLE IV 3a. Efficiency of metabolisable energy (M_{500} , kcal) for gain (G_{500} , kcal) and requirement of metabolisable energy for gain

TABLE IV 3b. Regression of metabolisable energy (M_{500} , kcal) required for maintenance on crude fibre (F_{500} , g) ingested, and the reverse

| Table IV 3a | | | | | | |
|-------------|--------------|------------|-------------|------------|--------------|-------------|
| Method | $1/b_{MG.F}$ | $b_{GM.F}$ | $b'_{GM.F}$ | $b_{MG.F}$ | $1/b_{GM.F}$ | $b'_{MG.F}$ |
| 1. | +0.6976 | +0.6224 | +0.6410 | +1.4335 | +1.6067 | +1.5601 |
| 2. | +0.6970 | +0.6284 | +0.6452 | +1.4348 | +1.5914 | +1.5499 |
| 3. | +0.7643 | +0.6725 | +0.7003 | +1.3085 | +1.4871 | +1.4279 |
| 4. | +0.7595 | +0.6748 | +0.6998 | +1.3166 | +1.4820 | +1.4290 |
| Average | +0.7296 | +0.6495 | +0.6716 | +1.3734 | +1.5418 | +1.4917 |

| Table IV 3b | | | | | | |
|-------------|--------------|------------|-------------|------------|--------------|-------------|
| Method | $1/b_{MF.G}$ | $b_{FM.G}$ | $b'_{FM.G}$ | $b_{MF.G}$ | $1/b_{FM.G}$ | $b'_{MF.G}$ |
| 1. | +1.6626 | +0.7891 | +1.3484 | +0.6015 | +1.2673 | +0.7416 |
| 2. | +1.6083 | +0.8223 | +1.3319 | +0.6218 | +1.2161 | +0.7508 |
| 3. | +2.4009 | +0.7440 | +1.8450 | +0.4165 | +1.3441 | +0.5420 |
| 4. | +2.2127 | +0.7901 | +1.7457 | +0.4519 | +1.2657 | +0.5729 |
| Average | +1.9711 | +0.7864 | +1.5678 | +0.5229 | +1.2733 | +0.6518 |

When the correlation is strictly linear the differences between $b_{MG.F}$ and $1/b_{GM.F}$ would decrease with increasing range of the variables G and M . With ever increasing range both values would converge to the same asymptotic limiting value lying somewhere in between.

There exists no reliable method to calculate an unbiased figure for this limiting value. Nevertheless we can calculate an average value by a method proposed many years ago by VAN UVEN (1927, 1930). This method consists in a modification of the method of least squares. Planes or hyperplanes of regression are calculated by minimising the sum of squares of the perpendiculars on these planes dropped from the points representing the observations. The difference with the ordinary method of least squares is that in the latter not the sum of the squares of perpendiculars is minimised but the sum of the squares of the distances parallel to one of the coordinates. VAN UVEN's principle is elucidated in fig. 1 in which only two variables have been considered.

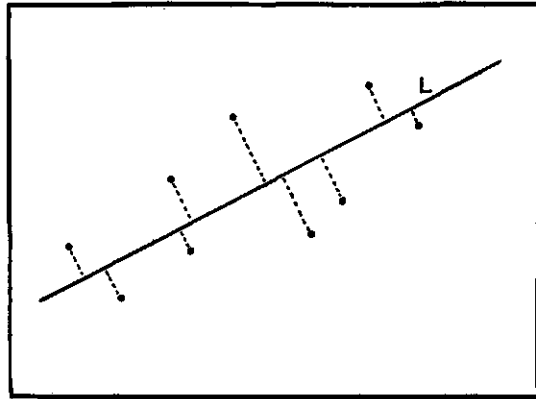


FIG. 1.
VAN UVEN's method of computing regression equations demonstrated with two variables. The sum of squares of the perpendiculars on the regression line dropped from the points representing the observations is minimised.

In our case three variables (M , G and F) had to be taken into account. In the calculation, again the general formula (IV 5) was the basis (22 M 's and 9 v 's). This formula was changed in the following way:

$$-I_{ij} - (-1)^{i+1}u_i + \alpha G_{500,ijt} + \beta(F_{500,ijt} - 1717) + \gamma M_{500,ijt} \sim 0 \quad (\text{IV } 8)$$

The unknowns are I_{ij} (22 items), u_i (9 items), α and β (one item each).

The following sum of 44 squares was minimised:

$$\sum \{-I_{ij} - (-1)^{i+1}u_i + \alpha G_{500,ijt} + \beta(F_{500,ijt} - 1717) + \gamma M_{500,ijt}\}^2,$$

with the condition

$$\alpha^2 + \beta^2 + \gamma^2 = 1.$$

The calculation finished, the formula was again rearranged into the formula (IV 5), so that

$$a = b'_{MG.F} = -\frac{\alpha}{\gamma}, \quad \frac{1}{a} = b'_{GM.F} = -\frac{\gamma}{\alpha}, \quad b = b'_{MF.G} = -\frac{\beta}{\gamma},$$

$$\frac{1}{b} = b'_{FM.G} = -\frac{\gamma}{\beta}, \quad M_{m,500,ij} = \frac{1}{\gamma} I_{ij}, \quad v_i = \frac{1}{\gamma} u_i.$$

The values of the partial regression ($b'_{GM,F}$) of G on M obtained in this way are inserted in the fourth column of table IV 3a. These values are indeed intermediate between those of the preceding two columns and would represent acceptable values for the efficiency of metabolisable energy for body gain.

In a still more refined calculation the fact that the experimental error of G is much higher than that of M could have been taken into account. In doing so $b'_{GM,F}$ would have shifted somewhat further to $1/b_{MG,F}$. This procedure would however have increased the labour of calculation without yielding appreciably more information.

The reciprocals $b'_{MG,F}$ are given in the last column of table IV 3a. As might be expected they are intermediate between the values $b_{MG,F}$ and $1/b_{GM,F}$ found in the preceding two columns.

A drawback of this modified method of least squares is that the regression coefficients are not essentially independent from the units in which the variables are expressed, in contradiction to the ordinary method of least squares. With this in mind our calculations of the regression coefficients with VAN UVEN'S method have been standardised by reducing the three variables, before the calculations, to equal standard deviation. This could easily be obtained by multiplying the variables with appropriate factors. Afterwards the variables and regression coefficients were transformed back into the original units.

In order to get an idea about the influence of this standardisation in some instances two values after the VAN UVEN method of least squares have been computed, one with reduction to standard deviation and one without such reduction. The regression coefficients were however only very slightly different, as may be seen from the following figures; $b'_{MG,F}$ and $b'_{MF,G}$ have been calculated with reduction to equal standard deviation of the variables and $b''_{MG,F}$ and $b''_{MF,G}$ without.

| Calculation | $b'_{MG,F}$ | $b''_{MG,F}$ | $b'_{MF,G}$ | $b''_{MF,G}$ |
|-------------|-------------|--------------|-------------|--------------|
| 1. | + 1.560 | + 1.587 | + 0.742 | + 0.784 |
| 2. | + 1.550 | + 1.574 | + 0.751 | + 0.789 |

In calculation 2 the figures for gain had been corrected to 12 hr standing, in calculation 1 they were uncorrected. The figures for gain (G) had not been multiplied by c_s/\bar{c}_s .

Information on the partial regression of M (metabolisable energy, kcal) on F (crude fibre, g) and the reverse are given in table IV 3b. It appears that the differences between $1/b_{MF,G}$ and $b_{FM,G}$ are much larger than those between $1/b_{MG,F}$ and $b_{GM,F}$ in the same columns of table IV 3a. The same applies to $b_{MF,G}$ and $1/b_{FM,G}$.

From this it can be concluded that the partial correlation between M and G

(gain, kcal) must be considerably higher than that between *M* and *F*. Therefore, all total and partial correlation coefficients resulting from the calculations with formula (IV 5) have been collected in table IV 4.

TABLE IV 4. Correlation coefficients

| Method | Total | | | Partial | | |
|--------|----------|----------|----------|------------|------------|------------|
| | r_{MG} | r_{MF} | r_{GF} | $r_{MG.F}$ | $r_{MF.G}$ | $r_{GF.M}$ |
| 1. | +0.919 | -0.495 | -0.737 | +0.945 | +0.689 | -0.826 |
| 2. | +0.921 | -0.495 | -0.741 | +0.950 | +0.715 | -0.841 |
| 3. | +0.932 | -0.495 | -0.688 | +0.938 | +0.557 | -0.720 |
| 4. | +0.932 | -0.495 | -0.697 | +0.943 | +0.598 | -0.750 |

Especially high are the partial correlations between *M* and *G*, i.e. $r_{MG.F}$ the average value is 0.944. Those between *M* and *F* and between *G* and *F* are only moderate. This means that the requirement of metabolisable energy (*M*) for gain (*G*) and the efficiency of metabolisable energy for gain (*G*) can be calculated from our data with much higher accuracy than the regressions between *M* and *F* and those between *G* and *F*.

This is somewhat surprising because in planning the experiments it has been the intention to study i.a. the interdependence of *M* and *F* while, in order to get a more clear cut result, *G* was kept as small as possible. In section IV.4 it will be shown that some spurious correlation may have increased the correlation between *M* and *G*.

Finally the regression formulae calculated with VAN UVEN's formulae with the four methods mentioned before (without and with multiplication of gain by c_s/\bar{c}_s , and without and with correction to 12 hr standing) are collected in table IV 5 and will be discussed more closely later on. All the values refer to 500 kg of live weight. $\bar{M}_{m,500}$ represents the average maintenance requirement (± 12000 kcal) with zero gain and ingestion of 1717 g of crude fibre a day. $\bar{M}_{m,500}$ is therefore the average of all values $M_{m,500,i} + (-1)^{i+1}v_i$.

TABLE IV 5. Regression of metabolisable energy (M_{500} , kcal) on gain (G_{500} , kcal) and on crude fibre (F_{500} , g) calculated with VAN UVEN's method. All values $\bar{M}_{m,500}$ are averages

| | |
|---------------------|--|
| Method 1: | $M_{500} = \bar{M}_{m,500} + 1.5601G_{500} + 0.7416(F_{500} - 1717)$ |
| Method 2: | $M_{500} = \bar{M}_{m,500} + 1.5499G_{500} + 0.7508(F_{500} - 1717)$ |
| Method 3: | $M_{500} = \bar{M}_{m,500} + 1.4279G_{500} + 0.5420(F_{500} - 1717)$ |
| Method 4: | $M_{500} = \bar{M}_{m,500} + 1.4290G_{500} + 0.5729(F_{500} - 1717)$ |
| Average | $M_{500} = \bar{M}_{m,500} + 1.4917G_{500} + 0.6518(F_{500} - 1717)$ |

TABLE IV.6. Metabolisable energy $M_{m,500,ij}$ (kcal) required for maintenance of 500 kg of live weight in the single experiments, calculated with VAN UVEN'S method (formula IV.5 and 8); zero balance and 1717 g crude fibre intake per 500 kg per day; method 4

| 1 Cow | 2 Single period | 3 Mtb. energy uncorrected but reduced to 500 kg l.w. | 4 Gain (kcal) uncorrected but reduced to 500 kg l.w. | 5 Mtb. energy corrected for gain (G) and crude fibre (F); 12 hr stand. | 6 Mtb. energy corrected for gain (G), crude fibre (F) and σ ; 12 hr stand. |
|-------------|--------------------|---|---|---|--|
| Zwartsch. 1 | R 17 | 13633 | + 815 | 12641 | 12382 |
| " | R 18 | 12546 | + 45 | 12099 | 12358 |
| Zwartkop 2 | R 17 | 12744 | - 244 | 12463 | 12722 |
| " | R 18 | 14439 | +1339 | 12958 | 12698 |
| Kee 2 | R 19 | 14646 | +1510 | 12444 | 12276 |
| " | R 20 | 12034 | + 20 | 11396 | 11564 |
| Alke | R 19 | 12967 | - 492 | 12760 | 12928 |
| " | R 20 | 14684 | +1484 | 12384 | 12216 |
| Kee 2 | R 25 | 14238 | +2530 | 11062 | 11220 |
| " | R 27 | 14077 | +1528 | 12286 | 12128 |
| Alke | R 25 | 13143 | +1802 | 11175 | 11017 |
| " | R 27 | 13909 | +1730 | 11768 | 11926 |
| RW 12 | R 29 | 12269 | + 242 | 12026 | 12220 |
| " | R 30 | 12679 | + 410 | 12088 | 11894 |
| Zwartsch. 2 | R 29 | 12394 | - 197 | 12988 | 12795 |
| " | R 30 | 12568 | + 702 | 12274 | 12468 |
| RW 12 | R 31 | 12448 | + 386 | 11942 | 11930 |
| " | R 32 | 13955 | + 845 | 11863 | 11875 |
| Zwartsch. 2 | R 31 | 14116 | +1157 | 11721 | 11732 |
| " | R 32 | 12585 | + 928 | 11688 | 11677 |
| Alke | R 33 | 11709 | + 936 | 11032 | 11346 |
| " | R 34 | 10892 | - 210 | 11118 | 10804 |
| RW 12 | R 33 | 11021 | - 452 | 11496 | 11369 |
| " | R 34 | 12506 | +1780 | 10701 | 10828 |
| Zwartsch. 2 | R 33 | 11556 | + 314 | 11335 | 11359 |
| " | R 34 | 12751 | +1874 | 10842 | 10818 |
| Kee 3 | R 33 | 13098 | + 938 | 12376 | 12166 |
| " | R 34 | 11580 | + 95 | 11413 | 11624 |
| Alke | R 35 | 10008 | - 483 | 10771 | 10916 |
| " | R 36 | 9737 | -1286 | 11354 | 11210 |
| RW 12 | R 35 | 10193 | - 634 | 10987 | 10937 |
| " | R 36 | 10931 | - 34 | 11180 | 11231 |
| Zwartsch. 2 | R 35 | 9935 | - 710 | 10994 | 11024 |
| " | R 36 | 11110 | - 7 | 11350 | 11319 |
| Kee 3 | R 35 | 10891 | - 282 | 11371 | 11246 |
| " | R 36 | 10169 | - 948 | 11416 | 11540 |
| Jansje | R 38 | 11785 | - 638 | 12927 | 13009 |
| " | R 39 | 10899 | -1436 | 12677 | 12594 |
| Roosje | R 38 | 11347 | -1787 | 13327 | 13245 |
| " | R 39 | 12068 | - 279 | 12748 | 12830 |
| Jansje | R 40 | 11750 | - 887 | 12704 | 12988 |
| " | R 41 | 10347 | -2455 | 13497 | 13213 |
| Roosje | R 40 | 10833 | -2079 | 13476 | 13192 |
| " | R 41 | 11620 | -1188 | 13135 | 13418 |

IV.2. MAINTENANCE REQUIREMENT

The corrected values $M_{500,ij}$ for the requirement of metabolisable energy per 500 kg live weight have been calculated for each cow in each single period with the aid of formula (IV 5). For the regression coefficients in this formula we substituted the values $a = b'_{MG,F} = +1.4290$ and $b = b'_{MF,G} = +0.5729$ from table IV 5, method 4; moreover the 9 values v_i were computed and taken into account. Since all four methods mentioned in table IV 5 essentially had given the same regression coefficients only one set of coefficients has been used, based on calculations with method 4 in which the figures for gain had been reduced to 12 hr standing and had been multiplied by c_s/\bar{c}_s ; negative balances had been multiplied by 0.83.

The results are given in table IV 6 in which the correction is performed in two steps. Column 3 and 4 contain the original data reduced to 500 kg live weight. The figures in column 5 are corrected for crude fibre and for gain, and those in column 6 are also corrected for v_i .

Table IV 7 contains the averages of the corrected values of each double experiment. In this table with double entry the figures are arranged after (double) periods (columns) and after individuals (rows). It appears that within the columns the variation is only small. Within the rows the fluctuations are markedly greater. We tried an analysis of variance. However, in this asymmetric table, the calculation of the residual variance is very time-consuming. Moreover, in our case, the ordinary analysis of variance does not reflect some theoretical considerations. The high figures in block R 38/39, R 40/41, Jansje, Roosje, for instance, can be explained either by high maintenance requirement of both cows or by a high period level in both double periods. In the ordinary

TABLE IV 7. Metabolisable energy $M_{m,500,ij}$ (kcal) required for maintenance of 500 kg live weight calculated with VAN UVEN's method (zero balance and 1717 g crude fibre intake per 500 kg per day) for all the cows and all the double periods. Calculation with formula (IV 5 and 8); correction 12 hr stand.; with c_s/\bar{c}_s

| Double periods | R 17/18 | R 19/20 | R 25/27 | R 29/30 | R 31/32 | R 33/34 | R 35/36 | R 38/39 | R 40/41 |
|----------------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| Zwartsch. 1 | 12370 | | | | | | | | |
| Zwartkop 2 | 12710 | | | | | | | | |
| Kee 2 | | 11920 | 11674 | | | | | | |
| Alke | | 12572 | 11471 | | | 11075 | 11063 | | |
| RW 12 | | | | 12057 | 11902 | 11098 | 11084 | | |
| Zwartsch. 2 | | | | 12631 | 11704 | 11088 | 11172 | | |
| Kee 3 | | | | | | 11895 | 11393 | | |
| Jansje | | | | | | | | 12802 | 13100 |
| Roosje | | | | | | | | 13037 | 13305 |
| Average | 12540 | 12246 | 11572 | 12344 | 11803 | 11289 | 11178 | 12920 | 13202 |

analysis of variance, the two possibilities are considered as equally probable. However, as a matter of fact, high period level is more probable, since a positive correlation between adjacent double periods is likely whereas it is probably absent between adjacent cows.

A split-up of the variance into two parts instead of three presents however no difficulties and nevertheless affords valuable information. At first we consider the columns and find

| | sum of squares |
|---------------------------|----------------|
| between columns | 10656867, |
| within columns | 1082158. |

Taking into account the available degrees of freedom (8 and 13) we find:

$$z_c = \frac{1}{2} \ln \frac{1332108}{83243} = 1.386.$$

From FISHER's table it follows that the fluctuations between columns, i.e. the period variations, cannot be considered as purely accidental (probability of being accidental < 0.1 %).

The same was done for the rows:

| | Sum of squares |
|------------------------|----------------|
| between rows | 7677881, |
| within rows | 4061144. |

$$z_r = \frac{1}{2} \ln \frac{959735}{312396} = 0.561.$$

Here the probability of being accidental was more than 5%.

Thus, significant variation between rows cannot be demonstrated in this way, but nevertheless could exist, since the variation within rows certainly would have been lower if period variation had been absent. The period variation, however, is quite clear. Therefore, the fluctuations of the $M_{m,500,ij}$ -values can be explained for a large part by period variation, that means without the hypothesis of individual variation, although such individual variations, of course, will not be completely absent.

Still more instructive is fig. 2. In this graph the *corrected* figures of the single experiments are plotted and the points referring to the same double experiment are connected by short lines. In every double period the lines are strictly parallel because v_i was supposed to be the same for all cows in double period i , but not in different double periods.

In different double periods the lines are lying on different levels between 11000 and 13000 kcals. Within the single double periods, however, they are lying quite close together, with only a few exceptions. These exceptions can

hardly be considered as being the result of between animal variation because the same pair of cows may behave different in different double periods. In double period R 19/20, for instance, the line of Alke is on a higher level than that of Kee 2; in double period R 25/27, however, the levels are reversed. The same was the case with cow Zwartschoft 2 and RW 12 in double period R 29/30 and R 31/32.

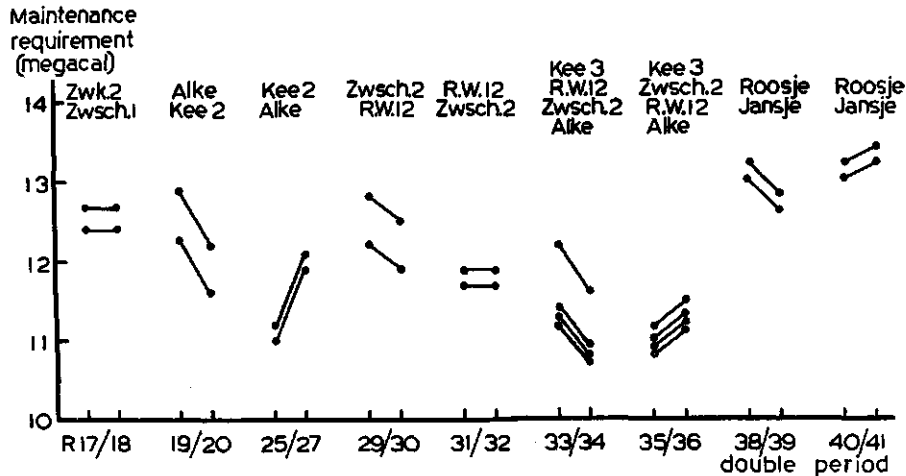


FIG. 2. Metabolisable energy required by single cows for maintenance in the two periods of the successive reversal trials: R 17/18, R 19/20 etc. The requirements are reduced to 500 kg live weight and 1717 g of crude fibre intake a day, and in the single reversal trials corrected to the same between period variation. In each reversal trial the results of the same cow in the two periods are connected by short lines. The names of the cows are given in the same order as the lines underneath. Note the large differences between the single reversal trials.

Also from this graph we must therefore conclude that, in these experiments, between double period variation is much more conspicuous than between animal variation. The causes of this between double period variation are not clear to us. The season might have some influence. Although always the same methods were used it is reasonable that also slight changes from period to period in the performance of measurements and in the analyses of gases, feed and excreta may have increased these between period variations. However, the errors produced in this way cannot be large because all determinations were performed with utmost care; gas measurements and gas analyses for example were checked several times. More important seems to be the fact that in a series of experiments heat expenditure often is higher in the first experiment than in the succeeding experiments (fig. 2; first experiments are performed in period R 19, R 29, R 33, R 38). In this article we will not go further into details. Of course, this phenomenon must have our full attention in the near future.

In table IV8 the corrected $M_{m,500,ijt}$ -values are tabulated in columns for early and late cut hay. As might be expected the difference between the early and late cut hay are very small since all intakes are corrected to zero balance and 1717 grams of crude fibre intake. Moreover the figures referring to the hays of different years are materially the same, the difference between the highest and lowest value being only 3 %. Nevertheless this result is not perfectly reliable, because the between double period variations are not eliminated.

TABLE IV8. Metabolisable energy $M_{m,500,ijt}$ (kcal) in early and late cut hays required for maintenance of 500 kg live weight, calculated with VAN UVEN'S method (formula IV 5 and 8; zero balance and 1717 g crude fibre per 500 kg per day)

| | 1958 early | 1958 late | 1959 early | 1959 late | 1960I early | 1960I late | 1960II early | 1960II late | Average |
|-------------|---------------|--------------|---------------|--------------|----------------|---------------|-----------------|----------------|---------|
| Zwartsch. 1 | 12382 | 12358 | | | | | | | 12370 |
| Zwartkop 2 | 12698 | 12722 | | | | | | | 12710 |
| Kee 2 | 12276 | 11564 | 12128 | 11220 | | | | | 11797 |
| Alke | 12216 | 12928 | 11017 | 11926 | 11346 | 10804 | 10916 | 11210 | 11545 |
| RW 12 | 11930 | 11875 | 12220 | 11894 | 10828 | 11369 | 11231 | 10937 | 11536 |
| Zwartsch. 2 | 11677 | 11732 | 12468 | 12795 | 10818 | 11359 | 11319 | 11024 | 11649 |
| Kee 3 | | | | | 12166 | 11624 | 11246 | 11540 | 11644 |
| Jansje | | | | | 13009 | 12594 | 12988 | 13213 | 12951 |
| Roosje | | | | | 12830 | 13245 | 13418 | 13192 | 13171 |
| Average | 12196 | 12196 | 11958 | 11959 | 11833 | 11832 | 11853 | 11853 | |

The fluctuation of the averages of the individual cows (table IV8) is much greater; the range is between 11536 and 13171 kcal, thus $\pm 13\%$. We have however already explained that the true between animal fluctuation probably is much smaller, the latter being disturbed by the between double period variations.

The best method for obtaining an average figure with standard deviation for maintenance requirement seems to calculate the non weighted average of the bottom row of table IV8. In this way we found that in our hays (33.5 % crude fibre in the organic matter; 49.1 % of the gross energy metabolisable) 12122 ± 710 kcal of metabolisable energy is needed for maintenance of a cow of 500 kg. The standard deviation refers to *one* animal in *one* double period. The average over all nine double periods is 12122 ± 237 kcal. Truly the standard deviation must be a little higher because the within column variation (between animal variation) should be included in this result. In doing so we found the following approximate values: 12122 ± 741 with average: 12122 ± 247 , thus essentially the same as before. This result is based on the calculation with method 4. The three other methods will give about the same result, thus approximately 12000 kcal with standard deviation for one cow in one double period ± 750 kcal.

We now will compare these results with *some data in the literature* about maintenance values and their period fluctuations.

Our value 12000 ± 750 reasonably compares with VAN ES' results (1961) in this laboratory. With cows in the seventh month of pregnancy he found 13000 kcal as the maintenance requirement of a cow of 500 kg and estimated the requirement of a non-pregnant cow of the same live weight to 12600 kcal on rations with on the average 27.85 % of crude fibre in the organic matter.

KELLNER (1900) determined the maintenance requirement of lean steers on rations of hay (grass hay or clover hay) and straw, or on grass hay or clover hay only. The live weight of the animals varied from 602 to 672 kg. Using MEEH's formula with power $2/3$ he estimated the requirement of an animal of 500 kg at 11 520 kcal a day.

This is in agreement with other results of VAN ES (1961) who established several regression formulae basing on European and American respiration trials with energy balances between +4000 and -4000 kcals. The average crude fibre content of the rations was 28.0 % on dry matter basis. From the first seven formulae referring to 64 experiments with *negative* balances the following maintenance requirements of metabolisable energy for 500 kg live weight may be computed: 11 420, 11 397, 11 417, 11 280, 11 422, 11 399 and 11 344 kcal respectively.

The first six formulae referring to 173 experiments with *positive* balances yielded 11 934, 11 830, 11 549, 11 000, 11 468 and 11 462 kcal. The average crude fibre content of the ration was 22.9 % in the dry matter.

These results agree with those of KELLNER just mentioned. In later experiments, however, KELLNER and FINGERLING obtained markedly higher results as may be computed from some other formulae of VAN ES referring to 69 respiration trials with positive balances of KELLNER and FINGERLING conducted on Bavarian steers with average live weight 695 kg. Calculated with the first eight of these formulae the maintenance requirement of animals with 500 kg of live weight proved to be 13 562, 12 984, 12 355, 11 800, 13 910, 13 932, 14 075 and 14 117 kcal. The average crude fibre content was 22.3 % in dry matter.

BREIREM (1944) pointed already out, many years ago, that the maintenance requirement computed from MØLLGAARD's experiments is markedly lower. VAN ES (1961) came to the same result. Six formulae derived from 27 Danish respiration trials with positive balances of MØLLGAARD in Red Danish cows (mean live weight 508 kg) gave the following results: 9 842, 9 810, 9 642, 9 949, 9 843 and 9 894 kcal per 500 kg of live weight. The average crude fibre content of the rations was 20.7 % on dry matter basis.

From these figures it follows that different laboratories have found largely different figures for maintenance requirement. This often has been thought to

be due to differences of breed. It is however also possible that period variation may intervene, because differences in maintenance requirement may occur in the same laboratory in different periods. This has already been described by BENEDICT et al. (1935) and was also shown by BREIREM (1944) in a statistical analysis of the Möckern experiments with Bavarian steers. Maintenance requirement was found markedly higher in the later experiments performed by FINGERLING than in the earlier ones of KELLNER. BREIREM supposed as a reason that the steers of FINGERLING were fatter than those of KELLNER.

Changes in maintenance requirement may even occur in short intervals. NEHRING et al. (1961) observed in their difference trials 'plötzliche Veränderungen' (sudden changes) in maintenance requirement of steers on a basal ration consisting of grass hay, clover hay and concentrates. With three animals the maintenance requirement in the first period with basal ration was 700 kcal, 1800 kcal and 1000 kcal higher than in a later period with the same basal ration. In another series of experiments the results from two out of eight animals had even to be dropped for this reason.

These changes are prejudicial to the determination of maintenance requirement. We reduced NEHRING's results with six animals for maintenance requirement (Versuchsreihe II) to 500 kg live weight in two ways: 1) by multiplication by $(500/W)^{3/4}$ and 2) by subtracting 13 kcal per kg of live weight over 500 kg. The results and standard deviations are: 11446 ± 1331 kcal and 12129 ± 1766 kcal respectively. These standard deviations are markedly higher than ours amounting to 741 kcal for between animal and between double period variation together.

In another set of experiments SCHIEMANN et al. (1961) fed four steers for almost one year on the same ration consisting of 4.3 kg of artificially dried hay, beet pulp and concentrates. Also in these experiments there were considerable within animal (between period) fluctuations of maintenance requirement in relatively short intervals. The standard between period deviation was 427 kcal when calculated from C-balances and 374 kcal when calculated from combustion values and gaseous exchange, thus somewhat smaller than in our experiments.

It is also reasonable that in long intervals the between period variation of maintenance requirement of metabolisable energy is greater than in short intervals. This follows from the statement of VAN ES (1961) that in experiments of MØLLGAARD and also in trials of this laboratory variation in respiratory exchange increases with increasing intervals between the determinations. Presumably the variation converges to a limiting asymptotic value.

The *character of the period variations* may be different. Firstly they may be *systematic* affecting all animals in a similar way as is suggested by our experiments (fig. 2). This was also seen in the investigation of SCHIEMANN et al.

(1961) with small animals: the responses of rabbits and rats were similar and depended on the season.

In other cases the period variations may be more *erratic*. This was observed with four steers in the just mentioned investigation of SCHIEMANN et al. The changes in metabolism did not go parallel; in one period the level of metabolism could be high in one animal and low in the other. The full data of the trials are however not published.

The between animal and between period variations not only make it difficult to determine average values for maintenance requirement, they also are prejudicial to the results of difference trials as in use for determining the fat production value of rations and single feeds. For obtaining reliable results *the design of difference trials* is of prime importance. Already the early investigators in this field eliminated between animal variation by feeding basal ration and experimental ration in each experiment to the same animal. Between period variation, however, has been thought to be duly allowed for by running one or two periods with basal ration, one before and (or) one after the period with experimental feed (two-period-trial and three-period-trial). In a longer lasting series of experiments the basal ration period following the first period with experimental feed is often also used for the next experimental period.

From a statistical point of view the results of such trials cannot be considered as independent in regard to systematic between period variation if two or more trials have one or more periods in common, that means when they are run at the same time, and also when the same basal ration period is used for two experiments. Such two- or three-period-trials only can be used safely for statistical work when they are not conducted at the same time and when they have no basal ration period in common. That means that the single two- or three-period-trials belonging to a set of such trials should follow each other with shorter or longer intervals.

| | Animal A | Animal B |
|---------------------|---|--|
| Preliminary period | Basal ration | Basal ration |
| <i>Fore period</i> | <i>Basal ration</i> fat prod.: A_1 | <i>Basal ration</i> fat prod.: B_1 |
| Transition period | Basal ration | Experimental ration |
| <i>Main period</i> | <i>Basal ration</i> fat prod.: A_2 | <i>Experimental ration</i> fat prod.: B_2 |
| Transition period | Basal ration | Basal ration |
| <i>After period</i> | <i>Basal ration</i> fat prod.: A_3 | <i>Basal ration</i> fat prod.: B_3 |

Statistically reliable results would however be obtained by running two trials simultaneously, one with only basal ration and the other with basal ration and experimental ration according to the following design.

For unbiased results the after period is not absolutely necessary but it increases the accuracy of the results. Without after period there remain four single experiments: three with basal ration and one with experimental ration.

In the way pointed out four or by preference six single experiments (determinations) are necessary for obtaining only one unbiased figure for fat production, three respectively five degrees of freedom being sacrificed.

The two formulae for the difference D in fat production between experimental ration and basal ration are respectively

$$D = B_2 - A_2 - (B_1 - A_1),$$

$$D = B_2 - A_2 - 1/2(B_1 - A_1 + B_3 - A_3).$$

In these formulae both, between animal variation and *systematic* between period variation, are eliminated. If σ_D stands for the *accidental* deviation of D and σ for the accidental deviation of A_1, A_2, B_1 etc. we have

$$\sigma_D^2 = 4\sigma^2 \quad \text{and} \quad \sigma_D^2 = 3\sigma^2$$

respectively.

The principle of these methods is the same as used in the Danish practical feeding trials with two or more groups of milch cows. These trials are run with many cows per group in one experiment. The same would be possible with the difference trials for determining feed value for fat production. This would leave the fat production values unbiased.

A still much better method of conducting difference trials furnishing unbiased results is the reversal method as used in our work. The design is as follows:

| | Animal A | Animal B |
|--------------------|---|---|
| Preliminary period | Basal ration | Experimental ration |
| Main period 1 | Basal ration fat prod.: A_1 | Experimental ration fat prod.: B_1 |
| Transition period | Experimental ration | Basal ration |
| Main period 2 | Experimental ration fat prod.: A_2 | Basal ration fat prod.: B_2 |

In this way two experiments with basal ration and two with experimental

ration, in total four single experiments, are necessary for obtaining only one unbiased figure for fat production value, three degrees of freedom being sacrificed. These can be considered as one for period variation between the first and second main period and two (for each animal one) for individual level of metabolism.

If the facilities allow to run two or three of such trials simultaneously the result is more favourable since it is reasonable to use for all cows only one degree of freedom for the variation between main period 1 and 2. Thus, for every pair of animals added to the first pair two more unbiased figures for fat production value become available.

For a judgement of the accuracy we consider the above design for two animals with two main periods. The result is:

$$D = 1/2(B_1 - A_1 + A_2 - B_2), \quad \text{with} \quad \sigma_D^2 = \sigma^2,$$

thus, much better than with the first design.

Of course it is possible to add a third transition and main period in which the rations are reversed for the second time (double reversal method). In this way there would be six single experiments with fat production $A_1, B_1, A_2, B_2, A_3, B_3$. The formulae are:

$$D = 1/4\{B_1 - A_1 + B_3 - A_3 + 2(A_2 - B_2)\}, \quad \text{with} \quad \sigma_D^2 = 3/4\sigma^2.$$

Three main periods, however, increase considerably duration, work and cost of the trial. On the other hand, reversal trials with only two main periods presumably will be quite satisfactory for most work on energy requirement for fat production. For a more extensive mathematical treatment of reversal trials (switch back trials) the reader should consult the papers of BRANDT (1938), SEATH (1944), TAYLOR et al. (1953) and LUCAS (1956).

From the above considerations it is clear that up to now figures for *maintenance requirement* may not always be reliable. The big between period variations should have the full attention of research workers in the near future.

It also follows that the feeding value of rations for maintenance can not yet be determined in a small number of one period trials as is done by determining digestion coefficients, that means by feeding a ration approximately providing for maintenance requirement so that afterwards only small corrections are necessary for obtaining the figure for the maintenance value of the ration.

If however a *reference feed* of constant maintenance value could be found difference trials would be possible also here for determining the ratio between the maintenance values of the experimental feed and the reference feed, both fed in quantities approximately meeting maintenance requirement. Again, unbiased results would be given by the reversal method which eliminates systematic between period variation and between animal variation. The design

would be the same as given higher up for reversal trials. Only the words 'basal ration' should be replaced by 'reference ration'.

In the present investigation the same principle has been followed. There was however not a constant reference feed but the two rations consisted of early and late cut hay of which every time one kind of hay or the other can be considered as reference feed.

IV.3. MAINTENANCE REQUIREMENT AND CRUDE FIBRE

In the preceeding paragraph it was shown that the mean value of the maintenance requirement is rather uncertain, probably as a result of the disturbing influence of the between period variations and between animal variations. Nevertheless, by elimination of these variations it is possible to study the increase and decrease of maintenance requirement as a function of crude fibre ingestion. We mentioned already that the ordinary regression coefficients obtained in this way are collected in table IV 1d. The average ordinary regression formula is

$$M_{500} = \bar{M}_{m,500} + 1.3734G_{500} + 0.5229(F_{500} - 1717).$$

If G_{500} is equal to zero M_{500} represents the maintenance requirement on a ration with F_{500} g crude fibre. The regression coefficient $b_{MF.G} = 0.5229$, therefore, can be interpreted as the increase of maintenance requirement per gram of crude fibre in the ration. We also could say that one gram of crude fibre ingested puts such a burden on metabolism that 0.5229 kcal of metabolisable energy is lost for maintenance purposes. The regression coefficient $b_{FM.G}$ of the inverse function can be interpreted as the grams of crude fibre metabolised per kcal of extra metabolisable energy. From other investigations it is highly probable that the extra heat is mainly the direct or indirect result of the rumen fermentations. It is not necessary to assume that this extra heat originates from degradation of crude fibre only.

The partial correlation coefficient between M_{500} and F_{500} (table IV 4) is not high. With the four methods outlined before we found 0.689, 0.715, 0.557 and 0.598. The average is 0.640. This low correlation is the reason that the differences between $b_{MF.G}$ and $1/b_{FM.G}$ are rather large, the averages being 0.523 and 1.273 respectively (table IV 3b).

Therefore, intermediate regression equations were calculated with VAN UVEN's formulae (table IV 5). The average is

$$M_{500} = \bar{M}_{m,500} + 1.4917G_{500} + 0.6518(F_{500} - 1717). \quad (IV 9)$$

The mean $b'_{MF.G}$ proved to be 0.652, a value not much higher than the average $b_{MF.G} = 0.523$ (table IV 3b).

In section IV.4 it will be explained that there exists some spurious correlation

between M and G . In that section new formulae are given after having eliminated spurious correlation as far as possible (table IV 1e and IV 10). The average ordinary regression equation (table IV 1e) proved to be

$$M_{500} = \bar{M}_{m,500} + 1.4391G_{500} + 0.5838(F_{500} - 1717).$$

With VAN UVEN's formulae (table IV 10) we found

$$M_{500} = \bar{M}_{m,500} + 1.5618G_{500} + 0.7160(F_{500} - 1717).$$

Comparison of this equation with (IV 9) shows an increase of the coefficient of $(F_{500} - 1717)$ from 0.652 to 0.716.

All these values are markedly lower than KELLNER's subtraction for crude fibre in fat production amounting to 1.36 NK_F per gram of crude fibre. KELLNER (1900) stated that one kg of digestible starch contains 3760 kcal of metabolisable energy. In his textbook (1905) he maintains that 1 kg digestible starch produces 248 g of fat with 9.5 kcal per gram, thus $248 \times 9.5 = 2356$ kcal, corresponding with an efficiency of 62.9 % of the metabolisable energy. About the same figure, 62.3 %, is given by HOFFMANN et al. (1962), also referring to KELLNER's results. In experiments with sheep JUCKER (1948) found 68.6 %. Newer extensive investigations were performed by HOFFMANN et al. (1962). In six trials with steers they found 64.1 % and in seven trials with wethers the same value. The subtraction of 1.36 NK_F per gram of crude fibre would therefore correspond with $1.36/0.63 = 2.2$ kcal of metabolisable energy, a figure about 3.4 times as high as our figure 0.652 and three times as high as our figure 0.716, both referring to maintenance.

In a separate calculation we determined the regression of metabolisable energy (M_{500}) on gain and percentage (f) of crude fibre in the organic matter with VAN UVEN's formulae. The result is given in table IV 9.

TABLE IV 9. Regression of metabolisable energy requirement (M_{500} , kcal) on gain (G_{500} , kcal) and on crude fibre percentage (f), calculated with VAN UVEN's method. All values $\bar{M}_{m,500}$ are averages

| | |
|---------------------|--|
| Method 1: | $M_{500} = \bar{M}_{m,500} + 1.7656G_{500} + 82.29(f-\bar{f})$ |
| Method 2: | $M_{500} = \bar{M}_{m,500} + 1.7569G_{500} + 83.48(f-\bar{f})$ |
| Method 3: | $M_{500} = \bar{M}_{m,500} + 1.5413G_{500} + 55.38(f-\bar{f})$ |
| Method 4: | $M_{500} = \bar{M}_{m,500} + 1.5518G_{500} + 59.16(f-\bar{f})$ |
| Average | $M_{500} = \bar{M}_{m,500} + 1.6539G_{500} + 70.08(f-\bar{f})$ |

The average is

$$M_{500} = \bar{M}_{m,500} + 1.6539G_{500} + 70.08(f-\bar{f}), \quad (IV 10)$$

in which $\bar{f} = 33.55$.

There is an increase of maintenance requirement with increasing crude fibre percentage in the organic matter amounting to 70 kcal per percent of crude

fibre. It should however be pointed out that this formula is only valid for small values of $(f - \bar{f})$, because the dependence of G_{500} on crude fibre content is not taken into account in the upper two formulae of table IV 9. In the lower two, however, allowance is made for this dependence.

Although the partial correlation between M_{500} and F_{500} proved to be rather low, we nevertheless are entitled to conclude that in hays more metabolisable energy is required for maintenance with higher crude fibre content of the ration.

This is at variance with the results of BLAXTER and his collaborators. From 15 experiments in which five foods were given to cattle and from 43 experiments in which 15 foods were given to sheep BLAXTER (1961) concluded that there was no association between the weight of dry food required for maintenance and the metabolisable energy required for maintenance. For widely different foods metabolisable energy was used with constant efficiency to meet the energy expenditure of maintenance. He maintained that a real difference exists in the way the energy of food is used to provide energy for doing work in the body, that is the muscular, osmotic and other work of maintenance, and the way it is used to promote synthesis of fat.

Since in BLAXTER's experiments there was no association between the weight of dry food required for maintenance and the metabolisable energy required for the same purpose, it probably follows that there neither would be an association between crude fibre ingested and metabolisable energy required for maintenance.

BLAXTER's results might also be tested by extending the formula (IV 5) in such a way that the coefficient of $G_{500,ijt}$ (gain) is not considered as a constant but dependent on F_{500} (crude fibre) and, moreover, different for positive and negative gain. In designating positive gain by G^+_{500} and negative gain by G^-_{500} , and $F_{500,ijt} - 1717$ by $F'_{500,ijt}$ we get:

$$M_{m,500,ij} + (-1)^{t+1}v_i + a_1(1 + \alpha_1 F'_{500,ijt})G^+_{500,ijt} + a_2(1 + \alpha_2 F'_{500,ijt})G^-_{500,ijt} + bF'_{500,ijt} = M_{500,ijt}$$

The number of unknowns is three more than in the formula (IV 5) used before. This makes the results uncertain, the more so because, in our experiments, G_{500} is only a correction factor. Nevertheless, a calculation was made. The result was however illogical as we had been afraid of.

As we found in our experiments a definite positive correlation between maintenance requirement and crude fibre we will briefly discuss whether the correction term with G_{500} (gain) in (IV 9) might obscure the true situation of things. Therefore, we remind that (IV 9) is the average of the four equations in table IV 5, in which G_{500} has four different meanings. In the upper two

equations the gains G_{500} have been used as such (uncorrected or corrected for standing and lying), except that negative balances have been multiplied by 0.83, thus not corrected for crude fibre. In the lower two G_{500} has been corrected for crude fibre by multiplication with c_s/\bar{c}_s , using KELLNER's formulae; see (IV2). It is true that these equations do not completely satisfy the deduction from BLAXTER's experiments that only requirement for positive gain is dependent on crude fibre ingestion in contradiction to negative gain. However, as correction with and without c_s/\bar{c}_s applied to G_{500} , gave essentially the same results, this objection cannot invalidate our results.

In our opinion the whole body of evidence including our experiments undoubtedly demonstrates that the requirement of metabolisable energy for maintenance is less dependent on crude fibre than requirement for fat synthesis. So far, however, a certain degree of dependence cannot be denied completely.

IV.4. METABOLISABLE ENERGY AND GAIN

We now will consider more closely the ordinary partial regression coefficients $b_{MG.F}$ of table IV1d referring to the requirement of metabolisable energy (M_{500} , kcal) for gain (G_{500} , kcal) with constant crude fibre intake (F_{500} , g). As mentioned, in the calculation of these regression coefficients (formula IV5) between period variation and between animal variation have been eliminated. The average value of $b_{MG.F}$ is +1.373. Furthermore, the mean $b_{GM.F}$ i.e. the efficiency of metabolisable energy for gain (table IV3a), is +0.650. Because of the high partial correlation ($r_{MG.F} = 0.94$) between M_{500} and G_{500} (table IV4) the reciprocal $1/b_{GM.F}$ of the efficiency, i.e. 1.542, is not much higher than $b_{MG.F} = 1.373$. The same applies to the average figures for the efficiency calculated as $b_{GM.F}$ and $1/b_{MG.F}$ being 0.650 and 0.730 respectively.

With VAN UVEN's formulae we calculated the intermediate regression formulae of table IV5 and the average formula:

$$M_{500} = \bar{M}_{m,500} + 1.4917G_{500} + 0.6518(F_{500} - 1717). \quad (\text{IV9})$$

The mean value of $b'_{MG.F}$ proved to be 1.492, corresponding with the efficiency: $1/b'_{MG.F} = b'_{GM.F} = 0.6704$, i.e. 67.0%. This figure refers to efficiency of metabolisable energy for gain with constant intake of 1717 g of crude fibre a day. Thus, in this formula increase of G_{500} is the result of increased intake of organic matter not containing crude fibre but mainly consisting of digestible carbohydrate. Our figure 67.0 should therefore be compared with the efficiency of starch for which KELLNER (1900, 1905) with steers, HOFFMANN et al. (1962) also with steers, and JUCKER (1948) with sheep found 62.6%, 64.1% and 68.6% respectively (see paragraph IV.3). Our result with cows is therefore somewhat higher than those of KELLNER and HOFFMANN et al. with steers; statistically the difference is not significant.

In hay the efficiency for gain found by KELLNER (1900) is much smaller. In four experiments with 'mittelgutes Wiesenheu' with 34.28 % crude fibre in the organic matter the efficiency was only 41.5 %. In our hays the crude fibre content was about the same: 33.55 %, again in the organic matter. Calculated with KELLNER's method the efficiency of our hays would only have been 46.2 %.

As a matter of fact these figures for efficiency referring to hay are not directly comparable with our figure 67.0 % mentioned above which supposes constant crude fibre intake. It is clear that in KELLNER's difference trials with hay crude fibre intake was different in the two periods of each difference trial as well as metabolisable energy intake, the ratio between these two remaining approximately constant. In our hays this ratio was 0.142 on an average.

We tried to recalculate the efficiency for gain of our hays in the following way starting from the average formula (IV9). As the ratio between crude fibre and metabolisable energy was 0.142, it follows that

$$(F_{500} - 1717) : (M_{500} - \bar{M}_{m,500}) = 0.1412.$$

Substitution in (IV9) gives

$$M_{500} - \bar{M}_{m,500} = 1.4917G_{500} + 0.6518 \times 0.1412(M_{500} - \bar{M}_{m,500}),$$

or

$$M_{500} = \bar{M}_{m,500} + \frac{1.4917}{1-0.0920} G_{500} = \bar{M}_{m,500} + 1.643G_{500}.$$

The result corresponds with an efficiency of 60.9 %. This was checked by calculations of the regression of metabolisable energy on gain and crude fibre percentage f using VAN UVEN's formulae. The results of the four methods of calculation have been collected in table IV9. The average formula, already mentioned before (IV10), is

$$M_{500} = \bar{M}_{m,500} + 1.6539G_{500} + 70.08(\bar{f}-f),$$

in which \bar{f} , the average crude fibre content of the organic matter, equals 33.55 %.

The requirement of metabolisable energy per kcal of gain proves to be 1.654 corresponding with an efficiency of 60.5 %. This agrees with the result obtained before (60.9 %).

This result is much higher than KELLNER's figures 41.5 and 46.2 mentioned before, and is not markedly lower than the figure for the efficiency of pure starch (ca. 63 %).

Still somewhat lower are the results obtained with a formula of BLAXTER (1961). BLAXTER studied the utilisation of the energy of food for lipogenesis in trials made above maintenance with 37 animals given 15 rations. Unfortunately, particulars about the species of the animals (presumably sheep) and rations are not given. For the efficiency (K) of metabolisable energy for fat

production (gain) the following formula was established:

$$K = 0.94 M - 8.0,$$

in which M stands for the metabolisability (%) of the gross energy.

In our hays M was 49.05 % so that the efficiency K would be 38.1, indeed somewhat lower than KELLNER's figures 41.5 and 46.2, and markedly lower than our figure 60.9.

More in line with our findings are the results of NEHRING et al. (1961). They conducted experiments with six steers on varying amounts of a ration of constant composition with 25.33 % crude fibre in the organic matter and consisting of artificially dried grass hay, artificially dried clover hay, beet pulp and concentrates. The efficiency of the metabolisable energy for fat production proved to be 57.7 % on an average, thus markedly higher than BLAXTER's and KELLNER's figures but still lower than our figure 60.9 for only grass hay with 33.55 % of crude fibre in the organic matter.

It is therefore doubtless that our figure is rather high and we looked for an explanation. In our experiments there were besides positive also several negative balances and in our computations they had been pooled after multiplication of the negative balances by 0.83. We mentioned already in paragraph IV.1 that according to BLAXTER's formulae the ratio would be only 0.50 instead of 0.83. We examined therefore whether our data would be better in agreement with another ratio than 0.83.

With this in mind we extended our formula (IV 5) in such a way that positive and negative balances were represented by separate terms. These balances had been corrected to 12 hour standing. Positive balances (kcal) are designated by G^+_{500} and negative ones by G^-_{500} . Again:

$M_{m,500,ij} + (-1)^{t+1}v_i$ = maintenance requirement of cow j in the first and second part ($t = 1$ or 2) of double period i , with crude fibre intake = 1717 g.

$G^+_{500,ijt}$ = positive gain (balance, kcal) of cow j in the first and second part ($t = 1$ or 2) of double period i , and similarly for $G^-_{500,ijt}$ (negative gain), for $F_{500,ijt}$ (grams of crude fibre ingested) and for $M_{500,ijt}$ (metabolisable energy ingested). All data refer to 500 kg live weight. a_1 , a_2 and b are constants:

$$M_{m,500,ijt} + (-1)^{t+1}v_i + a_1G^+_{500,ijt} + a_2G^-_{500,ijt} + b(F_{500,ijt} - 1717) = M_{500,ijt}.$$

Omitting the suffixes ijt , the solution for ordinary regression is

$$M_{500} = \bar{M}_{m,500} + 1.4467G^+_{500} + 1.1597G^-_{500} + 0.6159(F_{500} - 1717).$$

Two computations, with and without equalising standard deviations, with the aid of VAN UVEN's formulae gave:

$$M_{500} = \bar{M}_{m,500} + 1.5536G^+_{500} + 1.2650G^-_{500} + 0.7453(F_{500} - 1717),$$

$$M_{500} = \bar{M}_{m,500} + 1.5681G^+_{500} + 1.3469G^-_{500} + 0.7892(F_{500} - 1717).$$

We also calculated the reciprocals of the inverse regression coefficients:

$$a_1 = 1/b_{G^+M.G^-F} = 1.6282, \quad a_2 = 1/b_{G^-M.G^+M} = 1.5194.$$

The four ratio's a_2/a_1 are 0.802, 0.814, 0.859, and 0.933 respectively, thus quite near the MØLLGAARD value 0.83 used by us for pooling negative and positive balances in the earlier computations. The use in our calculations of an inappropriate ratio for the efficiency of metabolisable energy for positive gain and for prevention of negative gain is therefore eliminated.

The reason for the high efficiency of our hays for fat production is therefore not clear. One of the reasons might be that the balances were rather small or moderate at most. This had been done intentionally, because the main object was to determine maintenance requirement, so that it was advantageous to have only small gain corrections. It is however also possible that the efficiency of hay for gain is higher than generally believed, although presumably not as high as found by us up to now.

Still another reason might be some spurious correlation (BROUWER, 1944/1945). As a matter of fact metabolisable energy consumed (M_{500}) and balance (G_{500}) are not independent. Designating heat expenditure by H_{500} , we have

$$G_{500} = M_{500} - H_{500}.$$

If we put, while dropping the suffix 500:

$$M_i - \bar{M} = M'_i, \quad G_i - \bar{G} = G'_i, \quad H_i - \bar{H} = H'_i, \quad (i = 1, 2, \dots, n)$$

and $\frac{1}{n} \sum_{i=1}^n M_i'^2 = \overline{M'^2}$ etc. we find for the regression of M on G :

$$b_{MG} = \frac{\overline{M'G'}}{\overline{G'^2}} = \frac{\overline{M'(M' - H')}}{\overline{(M' - H')^2}} = \frac{\overline{M'^2} - \overline{M'H'}}{\overline{M'^2} - 2\overline{M'H'} + \overline{H'^2}}. \quad (\text{IV } 11)$$

Now we suppose for a while that M and H and therefore also M' and H' in (IV 11) have been determined without accidental analytical (and technical) errors. Then the introduction of the errors $\Delta M'$ and $\Delta H'$ would change the formula as follows. Assuming that $\Delta M'$ and $\Delta H'$ are independent and designating the estimates of their asymptotic standard values by $s_{M'}$ and $s_{H'}$ we find:

$$b^*_{MG} = \frac{\overline{M'^2} - \overline{M'H'} + s^2_{M'}}{\overline{M'^2} - 2\overline{M'H'} + \overline{H'^2} + s^2_{M'} + s^2_{H'}}.$$

Since $b_{MG} > 1$ and $s^2_{M'} < s^2_{M'} + s^2_{H'}$ we conclude that accidental independent errors of M' and H' decrease the value of b_{MG} . Thus, without these errors, requirement of metabolisable energy for gain would have been found higher and the efficiency $1/b_{MG}$ would have been found lower.

A similar result was found in studying the regression G on M . Here we found

$$b^*_{GM} = \frac{\overline{M'^2} - \overline{H'M'} + s^2_{M'}}{\overline{M'^2} + s^2_{M'}}$$

As $b_{GM} < 1$ and nominator and denominator are both increased by $s^2_{M'}$, it follows that without the accidental analytical (and technical) errors the efficiency b_{GM} would be smaller and the reciprocal $1/b_{GM}$ higher.

We also studied the effect of analytical errors on the partial regression coefficients $b_{MG.F}$ and $b_{GM.F}$ and found similar results. These results are also valid for our partial regression coefficients computed from the equations (IV 6).

In order to get some idea of the magnitude of this spurious correlation the calculations of the regression equations were repeated. In these new calculations (formula IV 5) the following values:

$$[MG] = [M(M - H)]; \quad [G^2] = [(M - H)^2]; \quad [M^2]$$

were reduced by n times the accidental variance of M , where n is the number of the experiments. Since the standard accidental error of M per single experiment had been found to be 1 % of the gross energy (section II.7.4), or ± 250 kcal per 500 kg body weight, the subtraction amounted to $n \times 250^2$.

The result is given in table IV 1e. The partial regression of M on G is indeed somewhat higher than in table IV 1d: on an average 1.44 instead of 1.37. The same applies to the regression of M on F : 0.58 instead of 0.52.

TABLE IV 10. Regression of metabolisable energy (M , kcal) on gain (G , kcal) and on crude fibre (F , g) calculated with VAN UVEN's method (formula IV 5 and IV 8); spurious correlation eliminated. All values $\overline{M}_{m,500}$ are averages

| | |
|---------------------|---|
| Method 1: | $M_{500} = \overline{M}_{m,500} + 1.6436G_{500} + 0.8182(F_{500} - 1717)$ |
| Method 2: | $M_{500} = \overline{M}_{m,500} + 1.6305G_{500} + 0.8256(F_{500} - 1717)$ |
| Method 3: | $M_{500} = \overline{M}_{m,500} + 1.4862G_{500} + 0.5943(F_{500} - 1717)$ |
| Method 4: | $M_{500} = \overline{M}_{m,500} + 1.4870G_{500} + 0.6258(F_{500} - 1717)$ |
| Average | $M_{500} = \overline{M}_{m,500} + 1.5618G_{500} + 0.7160(F_{500} - 1717)$ |

The results obtained with VAN UVEN's formulae are given in table IV 10. Also in these equations the coefficients of G_{500} and $(F_{500} - 1717)$ are a little higher than the corresponding values in table IV 5. The average is:

$$M_{500} = \overline{M}_{m,500} + 1.5618G_{500} + 0.7160(F_{500} - 1717).$$

With constant crude fibre intake the requirement per kcal of gain was 1.5618 kcal and, therefore, the efficiency for gain 64.0 %, corresponding to KELLNER's and HOFFMANN's above mentioned figures 62.6 % and 64.1 %. Just as in the beginning of this section we calculated the requirement for gain with constant crude fibre percentage of 33.55 %. We found:

$$M_{500} = \overline{M}_{m,500} + 1.7375G_{500}.$$

The result corresponds with an efficiency of 57.6 %, thus again somewhat lower than our former figure 60.9 but a great deal higher than KELLNER's and BLAXTER's values 41.5, 46.2 and 38.1 mentioned before. The agreement with the value 57.7 % of NEHRING et al. is however very good although it should be kept in mind that in NEHRING's investigation the crude fibre content of the ration was only 25.33 %.

It appears, therefore, that even after the elimination of spurious correlation, the reason of the high efficiency of our hays for fat production as compared with KELLNER's and BLAXTER's results is not clear. Obviously, more experiments with high hay rations will be necessary for clarification.

CHAPTER V

ANALYSES WITH CORRECTION FACTORS FOR ENERGY BALANCES FROM LITERATURE

V.1. INTRODUCTION

In chapter IV it was assumed that negative balances could be expressed in the same units as positive ones by multiplication by 0.83. Although a similar factor was found in a special computation (section IV.4) and also in a statistical analysis of experiments in the literature (section IV.1), BLAXTER's work (1961) suggested a considerably lower factor. Furthermore, all corrections for body weight were performed with the factor $(500/W)^{3/4}$. Other investigators, however, used lower or higher exponents. Finally a simplified regression equation $M = M_m + aG + bF$ was used.

It was shown that slight deviations of the above mentioned two constants a and b hardly can have a big influence on the results obtained for the maintenance value of hay.

This is however less certain for the results obtained for the relation between maintenance requirement and crude fibre and for the ratio between metabolisable energy and gain (lipogenesis). The latter ratio was indeed found rather small and, therefore, the efficiency for gain rather high. Calculations with more elaborate formulae were tried but the body of data was too small for obtaining a definite decision. Therefore, it appeared appropriate to use other correction factors, if available, derived from experiments of ourselves and of others. Care should however be taken not to disturb the homogeneity of the material which might produce erroneous results.

With the early and with the late cut hay 1959 a difference trial was performed, with one animal for each hay, giving some information on the efficiency of the utilisation of this hay for lipogenesis. Because of the high error innate to the result of a difference trial with only two animals the result was unfit to be used as a general correction figure for all the experiments.

Another way to estimate the efficiency of utilisation of metabolisable energy for gain or for prevention of negative balances consists in the calculation of the starch value according to KELLNER from the digested organic constituents and from crude fibre and by multiplying the result with 2365, i.e. the net energy of 1 kg digested starch, after which the efficiency immediately can be computed. In case of negative balances the efficiency might be increased by division by 0.83, a factor originating from MØLLGAARD (1929) and also used in chapter IV.

For positive energy balances also the equation of BLAXTER (1961) might be used:

$$K = 0.94M - 8.0,$$

where K stands for the efficiency (%) of the utilisation of metabolisable energy for lipogenesis and M for the metabolisable energy expressed as a percentage of gross energy. For negative balances BLAXTER (1961) found an efficiency of about 76 % for all kinds of rations.

Finally, the same efficiency values might be used as in earlier work (BROUWER et al., 1961) which are mainly based on results of regression equations of metabolisable energy on energy balance and the reverse, both computed from results given in the literature.

V.2. THE DIFFERENCE TRIAL

In period R 25 two animals were fed early (cow Alke) and late (cow Kee 2) cut hay 1959 and in the next period R 26 the rations were increased up to a point not far from maximum appetite. Table V1 gives some details of both experiments.

In each of these two difference trials (with cow Alke and cow Kee) M_i will stand for the metabolisable energy in the single trials ($i = 1$ and 2 in period R 25 and R 26 respectively), G_i for body gain (kcal) and W_i for body weight. The correction for increase of maintenance requirement between R 25 and R 26 is estimated at 17 or 24 kcal per kg of body weight increase corresponding to power $3/4$ or 1 in the formula for maintenance requirement: $M = 12000 (W/500)^p$ in which we took $W = 600$.

Then, the formula for the efficiency (E , %) of the utilisation of metabolisable energy for gain is

$$E = \frac{G_2 - G_1}{M_2 - M_1 - (17 \text{ or } 24)(W_2 - W_1)} \times 100. \quad (\text{V1})$$

The efficiency proved to be 48.9 for cow Alke and 41.9 for cow Kee, using the higher power for body weight increase, and 47.4 and 39.7 while using the lower one. These figures are close to those computed with KELLNER's and BLAXTER's formulae (section V.3), but they are markedly lower than those calculated in chapter IV (regression analysis).

We may try to estimate the standard deviation of these percentages. Equation (V1), by abbreviation, may be written:

$$E = \frac{G}{M}.$$

Thus,

$$\Delta E = \frac{M\Delta G - G\Delta M}{M^2}, \quad (\Delta E)^2 = \frac{M^2(\Delta G)^2 - 2MG\Delta G\Delta M + G^2(\Delta M)^2}{M^4}.$$

TABLE V.1. Some details of the difference trials in R. 25-R. 26

| Animal | Period | Body weight (kg) | Dry matter (g) | Gross energy (kcal) | Metabolisable energy | | Heat expend. from gas. exch. (kcal) | Protein balance (g) (6.25 × N) | Energy balance ¹⁾ (kcal) |
|----------|--------|------------------|----------------|---------------------|----------------------|----------------------|-------------------------------------|--------------------------------|-------------------------------------|
| | | | | | as such (kcal) | as % of gross energy | | | |
| Alke | R. 25 | 561 | 5791 | 25252 | 14328 | 56.7 | 12404 | +18 | +1964 |
| Alke | R. 26 | 591 | 8703 | 38122 | 21668 | 56.8 | 16563 | +82 | +5203 |
| Increase | | +30 | | | +7340 | | | | +3239 |
| Kee 2 | R. 25 | 611 | 6768 | 29595 | 16547 | 55.9 | 13462 | +26 | +2941 |
| Kee 2 | R. 26 | 660 | 10003 | 44088 | 23863 | 54.1 | 18309 | +36 | +5512 |
| Increase | | +49 | | | +7316 | | | | +2571 |

¹⁾ Average of energy balance computed from C- and N-balance, and energy balance computed from metabolisable energy, gaseous exchange and urine-N.

If, in addition to the just mentioned symbols, H_i stands for the heat expenditure and if $H = H_2 - H_1$, the asymptotic values $\langle \rangle$ are:

$$\begin{aligned} \langle (\Delta E)^2 \rangle &= \sigma_E^2, & \langle (\Delta G)^2 \rangle &= \sigma_G^2, & \langle (\Delta M)^2 \rangle &= \sigma_M^2, \\ \langle \Delta G \Delta M \rangle &= \langle (\Delta M - \Delta H) \Delta M \rangle &= \sigma_M^2. \end{aligned}$$

Therefore,

$$\sigma_E^2 = \frac{M^2 \sigma_G^2 - 2MG\sigma_M^2 + G^2\sigma_M^2}{M^4} = \frac{G^2}{M^2} \left\{ \frac{\sigma_G^2}{G^2} - 2 \frac{\sigma_M^2}{GM} + \frac{\sigma_M^2}{M^2} \right\}. \quad (V2)$$

Estimates s_M and s_G for σ_M and σ_G occurring in this formula were computed as follows.

In section II.7.4 we computed an estimate s_{M_i} for σ_{M_i} which proved to be one percent of the gross energy as well for single experiments as for double experiments.

From $G_i = M_i - H_i$ it follows

$$\sigma_{G_i}^2 = \sigma_{M_i}^2 + \sigma_{H_i}^2, \quad (V3)$$

if correlation between M_i and H_i is negligible as was the case in earlier experiments. We need therefore σ_{M_i} and σ_{H_i} .

In single experiments σ_{H_i} was estimated at 1% of the heat production; in longer lasting experiments σ_{H_i} is higher. This is caused by additional variance of heat expenditure between experiments. If between single experiment variation (within double period variation) was included s_{H_i} was somewhat below 2% of the maintenance requirement, thus about 200. s_{M_i} and s_{H_i} were substituted in (V3).

Next, neglecting variance due to the correction power for live weight increase, $s_G^2 = s_{G_1}^2 + s_{G_2}^2$ and $s_M^2 = s_{M_1}^2 + s_{M_2}^2$ were computed and substituted in formula (V2), so that for the efficiencies already mentioned in this section we computed

$$\begin{aligned} \text{Alke:} & \quad 48.9 \pm 5.6 \% \text{ and } 47.4 \pm 5.5 \%, \\ \text{Kee 2:} & \quad 41.9 \pm 7.0 \% \text{ and } 39.7 \pm 6.8 \%. \end{aligned}$$

The errors of the efficiency are rather high. A better result might have been attained if the energy balances in the first period (R 25) had been nearer to zero. It is clear, therefore, that the value of E as found should not be used for the whole material.

V.3. EFFICIENCY OF UTILISATION OF THE METABOLISABLE ENERGY FOR LIPOGENESIS AND FOR MAINTENANCE CALCULATED WITH METHODS GIVEN IN THE LITERATURE

In each experiment we computed the starch value of the ration with KELLNER's method with the only difference that $0.80 \times$ digested crude protein was substituted for $0.94 \times$ digested true protein. Thus,

starch units = $0.80 \times \text{g digested crude protein} + 1.91 \times \text{g digested crude fat} + 1.00 \times (\text{g digested crude fibre} + \text{g digested N-free extractives}) - 0.58 \times \text{g crude fibre}$.

This was multiplied by 2.365, the number of kcal net energy (NK_F) of 1 g digested starch. The result representing the total NK_F in the ration, was divided by the number of kcal of metabolisable energy and multiplied by 100 for obtaining the figure for the efficiency percentage of metabolisable energy for lipogenesis:

| Efficiency | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|------------|----------|------|----------|------|-----------|------|------------|------|
| | early | late | early | late | early | late | early | late |
| | 49 | 40 | 51 | 50 | 52 | 43 | 46 | 40 |

For negative balances these values might be divided by 0.83.

With the equation of BLAXTER (see chapter IV, sec. 4) we computed, again for lipogenesis, the following values for the efficiency of the metabolisable energy:

| Efficiency | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|------------|----------|------|----------|------|-----------|------|------------|------|
| | early | late | early | late | early | late | early | late |
| | 44 | 33 | 45 | 43 | 48 | 34 | 36 | 29 |

In case of negative balances for cattle a value of 76 % is given by BLAXTER (1961) irrespective of the composition of the feed (see section V.1).

Finally, the same values of efficiency might be used as in earlier work mainly based on regression of energy balance on metabolisable energy and of metabolisable energy on energy balance in a large number of experiments of MØLLGAARD, KELLNER, FINGERLING, ARMSBY and FORBES with energy balances between +4000 and -4000 kcal (BROUWER et al., 1961; VAN ES, 1961). These were efficiencies of 50 % and 59 % for positive balances and 1/0.83 times as much for negative balances.

These figures show that there are considerable differences between the measured or estimated efficiencies. In general KELLNER's method has given values between 40 and 50 which are higher than those of BLAXTER's equation.

As to the values for the negative balances, those obtained with KELLNER's method are low and the BLAXTER-values are rather high.

The two values of the difference trial with early and late cut hay 1959 are close to those computed with BLAXTER's equation and not far from those computed with KELLNER's method.

Therefore, in the next section the maintenance requirement was computed

with several of the efficiency values recorded above in order to examine to what extent their use affects the maintenance requirement of metabolisable energy.

V.4. THE MAINTENANCE REQUIREMENT OF METABOLISABLE ENERGY PER 500 KG BODY WEIGHT ($M_{m,500}$) COMPUTED WITH VARIOUS CORRECTIONS FOR ENERGY BALANCE AND BODY WEIGHT

In all 44 single experiments we computed the maintenance requirement of metabolisable energy ($M_{m,w}$, kcal, where W = kg body weight) and reduced it then to 500 kg body weight ($M_{m,500}$, kcal). The correction for energy balance was done with most of the efficiency values of section V.3. The correction for body weight was made with the formula $M_{m,500} = (500/W)^p M_{m,w}$, using a value for p of 3/4 or, occasionally, of 1.

In addition, also the requirement of (computed) starch units ($S_{m,500}$) was calculated. With this purpose the content of starch units of each ration (see section V.3) was corrected for energy balance assuming that one starch unit would give a positive energy balance of 2.365 kcal or would prevent a negative balance of $2.365/0.83 = 2.849$ kcal. The correction for body weight was equal to the one of $M_{m,w}$ just mentioned.

We chose five combinations of the exponent p and the efficiencies of sec. V.3; they are given in table V2, first and second column. With each of these combinations a set of values $M_{m,500}$ for corrected metabolisable energy was computed, one value for each single experiment, thus 44 in total.

With each of these five sets analyses of variance were established for the 4 days 1958, 1959, 1960I and 1960II separately. In each of these four analyses of

TABLE V2. Analysis of variance of $M_{m,500}$ and $S_{m,500}$ computed with various gain and weight correct

| Efficiency used in correcting energy balance | Exponent used in weight correction p | Hay 1958 | | | | Hay 1959 | | | |
|--|--|---------------------|----------------------|----------------------------|------------------------------------|---------------------|----------------------|----------------------------|------------------------------------|
| | | F - value | | Residual variance d.f. = 5 | Average $M_{m,500}$ or $S_{m,500}$ | F - value | | Residual variance d.f. = 3 | Average $M_{m,500}$ or $S_{m,500}$ |
| | | Betw. days d.f. = 1 | Betw. anim. d.f. = 5 | | | Betw. days d.f. = 1 | Betw. anim. d.f. = 3 | | |
| Earlier work (59%) | 3/4 | 9* | 2 | 164730 | 12254 | < 1 | 2 | 787041 | 1128! |
| Earlier work (50%) | 3/4 | 14* | 3 | 186006 | 12051 | < 1 | 2 | 1088557 | 1095! |
| Earlier work (50%) | 1 | 11* | 4 | 172314 | 11738 | < 1 | 3 | 914622 | 1055! |
| KELLNER's method | 3/4 | 7* | 3 | 290820 | 11936 | < 1 | 2 | 1140349 | 1098! |
| BLAXTER | 3/4 | 5 | 2 | 433417 | 11739 | < 1 | 2 | 1452698 | 1064! |
| Starch equivalent | 3/4 | 16** | 4 | 13091 | 2243 | < 1 | 2 | 60476 | 235! |
| Starch equivalent | 1 | 21** | 6* | 11301 | 2187 | < 1 | 2 | 51457 | 227! |

* and **: significant at 5% and 1% level respectively
d.f. = degrees of freedom

variance the variance was partitioned in three parts. One part refers to variance between animals (four to six individuals). In this part between double period variance is included, because for each of these animals only two experiments (one experiment with early cut hay and one with late cut hay) always fell in one double period. The second part of the variance was for between early and late hay variance and the third part was the residual variance.

Within double period (between single period) variation was not accounted for, which has increased somewhat the residual variance. Presumably it also might have been possible to compute *one* residual variance (instead of four) with increased accuracy from each of the seven complete sets of 44 values for corrected metabolisable energy requirement, thus for all eight kinds of hay together.

Significant variance of $M_{m,500}$ between early and late cut hay was found with the hays 1958, 1960I and 1960II and between animal plus between double period variance for the hays 1960I and 1960II, almost regardless of the way in which $M_{m,500}$ was computed (table V2).

We also computed in each of the sets for each hay the average $M_{m,500}$ or $S_{m,500}$, i.e. the average requirement of the six or four animals used, and furthermore the difference of these averages for early and late hay of the same field (table V3, lower part). The standard error of this difference was computed from the differences for the four or six single animals fed with these hays.

From table V3 it may be seen that the use of the exponent 1 instead of 3/4 in the correction for body weight decreased all values $M_{m,500}$ and $S_{m,500}$ with 2-4%. Furthermore it decreased the differences of these values between early and late hay with about 1% of average $M_{m,500}$ or $S_{m,500}$.

A greater influence on the values $M_{m,500}$ as well as on the differences of these values between early and late hay had the use of the various values of efficiency

DRS

| Hay 1960 I | | | | Hay 1960 II | | | | Efficiency used in correcting energy balance |
|--------------------------|----------------------------|-------------------------------|---|---------------------------|----------------------------|-------------------------------|---|--|
| F - value | | Residual variance d.f. = 5 | Average $M_{m,500}$ or $S_{m,500}$ | F - value | | Residual variance d.f. = 5 | Average $M_{m,500}$ or $S_{m,500}$ | |
| etw. hays d.f. = 1 | Betw. anim. d.f. = 5 | | | Betw. hays d.f. = 1 | Betw. anim. d.f. = 5 | | | |
| 9* | 9* | 351 781 | 11 491 | 5 | 35** | 83 425 | 11 918 | Earlier work (59%) |
| 12* | 9* | 490 206 | 11 492 | 10* | 33** | 110 443 | 12 146 | Earlier work (50%) |
| 10* | 9* | 478 583 | 11 059 | 7* | 31** | 113 925 | 11 766 | Earlier work (50%) |
| 14* | 10* | 483 104 | 11 526 | 16** | 20** | 227 763 | 12 459 | KELLNER's method |
| 9* | 7* | 513 234 | 11 234 | 4 | 36** | 74 583 | 11 826 | BLAXTER |
| 2 | 11** | 20 233 | 2 297 | 7* | 29** | 5 842 | 2 255 | Starch equivalent |
| 3 | 11** | 20 119 | 2 222 | 8* | 27** | 6 048 | 2 185 | Starch equivalent |

of utilisation of metabolisable energy in correcting for energy balances. The lowest values of $M_{m,500}$ were for all hays obtained with the efficiency values of BLAXTER. The highest values were computed with the high efficiency value of the earlier work, especially in the experiments with mainly positive balances (1958, 1959 and 1960I early) whereas the efficiency values of KELLNER's method gave the highest values in the other experiments with mainly negative balances (1960I late, and 1960II early and late). The difference between highest and lowest values was 2-9 % of the average $M_{m,500}$.

The differences between the values $M_{m,500}$ of early and late hay as a percentage of average $M_{m,500}$, varied, due to the various efficiency values, from 6 to 8 % for hay 1958, from 0 to 2 % for hay 1959, from 9 to 13 % for hay 1960I and from 3 to 9 % for hay 1960II.

The differences of average $M_{m,500}$ between early and late hay were significant for the hays 1958, 1960I and 1960II for almost all efficiency values. Obviously the metabolisable energy requirement for maintenance was not equal for all hays. A similar conclusion was drawn from the results of the regression analysis.

We now make a comparison between the differences of $M_{m,500}$ of early and late hay as a percentage of average $M_{m,500}$ from table V3 on the one hand, with the differences which might have been expected if the metabolisable energy had been utilised with the same efficiency as for fat production (section V.3) on the other hand. This efficiency for fat production was computed with KELLNER's method or BLAXTER's equation.

| Efficiency computed with | | Difference of $M_{m,500}$ between early and late hay (% of average) | | | |
|--------------------------|---------------------|---|----------|-----------|------------|
| | | Hay 1958 | Hay 1959 | Hay 1960I | Hay 1960II |
| KELLNER's method | Found (table V3) | 7 | 1 | 13 | 9 |
| | Computed (sec. V.3) | 20 | 2 | 19 | 14 |
| BLAXTER's equation | Found (table V3) | 8 | 0 | 11 | 3 |
| | Computed (sec. V.3) | 29 | 4 | 34 | 22 |

The differences in $M_{m,500}$ as found (table V3) are much smaller than those computed in the two manners based on efficiency for fat production. This explains why the values $S_{m,500}$ for starch units of table V3 are not equal for the early and late hays of the same fields because these $S_{m,500}$ -values are also based on efficiency for fat production. Obviously, the efficiency of utilisation of metabolisable energy for maintenance varies considerably less than that for fat synthesis.

The differences in $M_{m,500}$ between early and late hay (table V3, lower part), found by correcting for negative and positive balances with the five methods

TABLE V3. Average maintenance requirement of metabolisable energy per 500 kg body weight computed with various corrections for energy balance and for body weight

| Efficiency used in correcting energy balance | Exponent used in correcting for body weight | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|--|---|--------------------------------|-------|----------|-------|-----------|-------|------------|-------|
| | | Early | Late | Early | Late | Early | Late | Early | Late |
| | | $M_{m,500}$ Earlier work, high | 11909 | 12600 | 11189 | 11389 | 10968 | 12013 | 11726 |
| $M_{m,500}$ Earlier work, low | 11585 | 12518 | 10867 | 11051 | 10730 | 12154 | 11846 | 12447 | |
| $M_{m,500}$ Earlier work, low | 1 | 11348 | 12128 | 10486 | 10622 | 10437 | 11682 | 11511 | 12022 |
| $M_{m,500}$ KELLNER's method | 3/4 | 11535 | 12336 | 10931 | 11044 | 10770 | 12282 | 11915 | 13002 |
| $M_{m,500}$ BLAXTER's equation and value | 3/4 | 11295 | 12184 | 10643 | 10649 | 10603 | 11864 | 11678 | 11974 |
| $S_{m,500}$ = Starch units | 3/4 | 2377 | 2110 | 2374 | 2344 | 2360 | 2235 | 2312 | 2198 |
| $S_{m,500}$ = Starch units | 1 | 2328 | 2045 | 2292 | 2253 | 2296 | 2148 | 2247 | 2123 |

Mean difference in $M_{m,500}$ or $S_{m,500}$ between early and late hay in kcal and as a % of average $M_{m,500}$ or $S_{m,500}$ and standard error of mean difference (late minus early)

| | Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|-----------------------------|--------------------|-----------------|-------------------|------------------|----------------|------|------------|------|
| | Early | Late | Early | Late | Early | Late | Early | Late |
| | Earlier work, high | 691* (6%) ± 234 | 200 (2%) ± 625 | 1044* (9%) ± 342 | 383 (3%) ± 166 | | | |
| Earlier work, low | 932* (8%) ± 249 | 184 (2%) ± 740 | 1424* (12%) ± 404 | 601* (5%) ± 192 | | | | |
| Earlier work, low | 780* (7%) ± 239 | 136 (1%) ± 676 | 1245* (11%) ± 399 | 511* (4%) ± 195 | | | | |
| KELLNER's method | 801* (7%) ± 311 | 113 (1%) ± 755 | 1512* (13%) ± 401 | 1087* (9%) ± 275 | | | | |
| BLAXTER's equation or value | 889 (8%) ± 380 | 6 (0%) ± 852 | 1260* (11%) ± 413 | 296 (3%) ± 158 | | | | |
| $S_{m,500}$ = Starch units | -268** (-12%) ± 66 | -31 (-1%) ± 174 | -125 (-5%) ± 82 | -114 (-5%) ± 44 | | | | |
| $S_{m,500}$ = Starch units | -283** (-13%) ± 61 | -39 (-2%) ± 160 | -148 (-7%) ± 82 | -124* (-6%) ± 45 | | | | |

* and ** = significant at 5 and 1% level of probability

mentioned in the first column of the upper part of this table, had about the same size for the hays 1958, 1959 and 1960I, which mainly had shown positive balances. For the hays 1960II the difference calculated with KELLNER's corrections was considerably larger than that computed with the other four methods. In all experiments with these hays 1960II, however, the balances had been negative. As in the correction with the KELLNER method the efficiency percentage is not constant but depends mainly on the crude fibre content in distinction from the other four methods it is probable that the KELLNER correction is not appropriate for correction for negative balances.

The average differences of maintenance value between the early and late hays 1958, 1959, 1960I and 1960II computed from the figures in table V3, lower part, are 7, 1, 11 and 5 % respectively. For further calculations the values 7, 1, 12 and 5 % were chosen because slightly more weight was given to the results obtained with the efficiencies of KELLNER and of BLAXTER. We compared these differences in $M_{m,500}$ with the differences in metabolisability, digestibility and crude fibre content of the hays.

| | Hay 1958 | Hay 1959 | Hay 1960I | Hay 1960II |
|--|-------------|-------------|--------------|---------------|
| Probable difference in $M_{m,500}$ ¹⁾ | 7 % | 1 % | 12 % | 5 % |
| Difference in metabolisability ²⁾ | -11.6 % | -2.6 % | -14.3 % | -7.8 % |
| „ „ digestibility of dry matter | -12.8 % | -3.0 % | -17.7 % | -10.5 % |
| „ „ digestibility of energy | -15.2 % | -3.8 % | -18.4 % | -10.7 % |
| „ „ crude fibre content of dry matter | 10.4 % | 3.3 % | 12.2 % | 4.4 % |

1) as a percentage of average $M_{m,500}$.

2) metabolisable energy as a percentage of gross energy.

There was a strong correlation between the probable differences in $M_{m,500}$ and either the metabolisability ($r = -0.97^*$), the digestibility of dry matter and of energy ($r = -0.98^*$ and -0.97^* respectively) or the crude fibre content ($r = +0.92$) where the asterisks mean significance. An increase of 1.45 unit of metabolisability, of 1.8 and 1.9 unit of the digestibility of dry matter and of energy and furthermore a decrease of 1.2 unit of the crude fibre content decreased the maintenance requirement with about one percent. BREIREM (1944, p. 15) computed a decrease of the maintenance requirement of the same size (1%) for a decrease of about 1.8 unit of the crude fibre content in 82 experiments with small energy balances published by ARMSBY et al. and FORBES et al.

For the whole material we computed within the double periods the regression of $M_{m,500}$, obtained in each of three ways in correcting for energy balance, on crude fibre ingested (F , g) and also on the percentage of crude fibre in the dry matter (f).

TABLE V4. Regressions

| Efficiency used to compute $M_{m,500}$ | Correction for variation within double periods | Regression equation | s |
|---|--|---|------|
| KELLNER's method (pos. bal. 40-50 %; neg. bal. 48-60 %) | not applied | $M_{m,500} = \bar{M}_{m,500} + (1.19 \pm 0.27) (F - \bar{F})$ | 1021 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (1.17 \pm 0.29) (F - \bar{F})$ | 755 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (111 \pm 23) (f - \bar{f})$ | 679 |
| BLAXIER's equation (pos. bal. 29-48 %; neg. bal. 76 %) | not applied | $M_{m,500} = \bar{M}_{m,500} + (0.93 \pm 0.27) (F - \bar{F})$ | 1017 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (0.91 \pm 0.28) (F - \bar{F})$ | 744 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (91 \pm 22) (f - \bar{f})$ | 642 |
| Earlier work, high values (pos. bal. 59 %; neg. bal. 71 %) | not applied | $M_{m,500} = \bar{M}_{m,500} + (0.87 \pm 0.19) (F - \bar{F})$ | 713 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (0.86 \pm 0.15) (F - \bar{F})$ | 396 |
| | applied | $M_{m,500} = \bar{M}_{m,500} + (78 \pm 13) (f - \bar{f})$ | 381 |

The regression coefficients are significant. Those of $F - \bar{F}$ are low compared with the value 2.2 derived earlier from KELLNER's crude fibre correction for fat production and higher than the coefficients found in the regression computations of section IV.3 in which the efficiency of the utilisation of the metabolisable energy for fat synthesis has been found to be very high (table IV 3a, b). The results show that the use of a higher value of efficiency in correcting for energy balance gives lower regression coefficients of $(F - \bar{F})$. The regression coefficients of $(f - \bar{f})$ are in agreement with the results derived above from correlation between the probable differences in $M_{m,500}$ and the crude fibre content, and also in agreement with table IV 9.

V.5. THE STANDARD DEVIATION OF $M_{m,500}$ IN A SINGLE EXPERIMENT

As already mentioned the difference of $M_{m,500}$ between early and late hay from each of the four fields was determined four or six times in reversal trials, each time with another animal. Thus, between animal and between double period variance had been eliminated, whereas within double period variation, being only moderate, had been neglected. From these differences the standard deviation of one single experiment was calculated by multiplying the standard deviations of the four mean differences with $\sqrt{n/2}$, n being the number of the differences per field.

In this way the standard deviations of the single experiments with the hays of 1958, 1959, 1960I and 1960II proved to be about $300\sqrt{3} = 500$, $800\sqrt{2} =$

= 1100, $400\sqrt{3} = 700$ and $180\sqrt{3} = 300$ kcal respectively, which is equal to 4, 10, 6 and 3 % respectively of the average $M_{m,500}$ of these hays. The same standard deviations were found in the analysis of variance of the values $M_{m,500}$ as might be expected (table V2, square root of residual variance).

In earlier work (VAN ES, 1961) a formula was derived to estimate the standard deviation ($s_{M_{m,500}}$) of $M_{m,500}$ in one single experiment from a) the standard deviations (in all the experiments) of metabolisable energy (s_M , kcal), b) that of heat production (s_H , kcal) and c) that of the reciprocal of the efficiency with which the metabolisable energy was utilised to prevent negative balances or to synthesise fat (s_c), together with the reciprocal (c) of the efficiency itself, and the average of the sum of squares of the energy balances in the experiments ($(1/n)\sum G^2$):

$$s_{M_{m,500}} = \sqrt{(c-1)^2 s_M^2 + (1/n) s_c^2 \sum G^2 + c^2 s_H^2}$$

In sections II.7.4 and II.7.5 of the methodical chapter we have found $s_M = 270$ kcal and s_H within a single experiment with six respiration days about 130 kcal, corresponding with 250 kcal and 120 kcal per 500 kg live weight. The values of $(1/n)\sum G^2$ were computed from the figures for energy balance (G) which had been corrected to 500 kg body weight. They proved to be 860000, 1970000, 1210000 and 1360000 for the hays 1958, 1959, 1960I and 1960II respectively.

For c a value of 1.5, 2.0 or 2.5 (efficiency 67, 50 or 40 %) might be used in experiments with positive balances (hays 1958, 1959) and a value of 1.5 (efficiency 67 %) in the experiments with negative balances (half of the experiments with hays 1960I and all experiments with hays 1960II).

For experiments with negative balances s_c is probably low since we found only small variations in maintenance requirement of metabolisable energy. We might therefore use a value 0.1 in which case in 2/3 of the experiments with negative balances the efficiency would have been between 62 and 71 %.

For the experiments with positive balances it is even more difficult to estimate s_c because of the varying efficiencies found by KELLNER, BLAXTER and in our section V.3. A value 0.3 probably does not underestimate s_c .

Calculated with the three c -values 2.5, 2.0 and 1.5 for positive and 1.5 for negative gain, and using $s_c = 0.3$ and 0.1 for positive and negative gain, respectively, the standard deviations $s_{M_{m,500}}$ expressed as a percentage of $M_{m,500}$ (coefficients of variation) were as follows:

| | Hay 1958 | Hay 1959 | Hay 1960I | Hay 1960II |
|--------------------------|----------|----------|-----------|------------|
| $c = 2.5$ | 5 | 6 | | |
| $c = 2.0$ | 4 | 5 | | |
| $c = 1.5$ | 3 | 4 | 4 | 2 |
| Actually found | 4 | 10 | 6 | 3 |

The values actually found higher up in this section are again given in the bottom line. The high deviation in the experiments with hay 1959 may have been due to mastitis from which three of the animals suffered in the preliminary period, but perhaps also in the experimental period although their temperature was normal.

Most of the values in the bottom line are a little higher than those in the other lines, presumably due to within double period (between single period) variation which is included in the figures in the bottom line in contrast to those in the other lines. The existence of such within double period variation of $M_{m,500}$ has been shown in chapter IV. It depends presumably on a higher variation of heat production between the two single experiments in a double period than within single experiments. Earlier results also showed a tendency in that direction (VAN ES et al., 1961). Our coefficient of variation of heat production for one respiration day in single periods of 2.5 % (section II.7.5) is rather low. Using the same animals fed with the same ration for about nine months SCHIEMANN et al. (1961) found between periods a coefficient of variation of about 4 % for one day.

V.6. THE MAINTENANCE REQUIREMENT OF ABSORBED ENERGY MINUS ENERGY IN URINE

Not all of the absorbed energy can be utilised by the animals if they are in the zone of thermoneutrality. The heat produced in the rumen is useless. From MARSTON's *in vitro* experiments BLAXTER (1961) has estimated the fermentation heat at 80 % of the energy in methane. Although this result only can be tentative for the living animal we computed the amount of 'corrected metabolisable energy' (metabolisable energy minus fermentation heat) as a percentage of the metabolisable energy.

| Hay 1958 | | Hay 1959 | | Hay 1960I | | Hay 1960II | |
|----------|--------|----------|--------|-----------|--------|------------|--------|
| early | late | early | late | early | late | early | late |
| 87.7 % | 85.6 % | 88.2 % | 87.8 % | 88.8 % | 86.8 % | 87.2 % | 86.2 % |

In this table the percentages for the early hay all are higher than for the late hay. Thus, we may conclude that the heat in methane and presumably also the fermentation heat are both higher on the late hay if calculated as a percentage of the metabolisable energy. However, since the metabolisable energy content of the late hay was considerably lower (table III 2), the percentages of heat in methane and of fermentation heat are also lower if calculated not on metabolisable energy basis but on gross energy basis, at least if BLAXTER's above mentioned estimation is correct.

After converting our figures for maintenance requirement of metabolisable

energy into figures for maintenance requirement of corrected metabolisable energy the differences between figures for early and late hay become somewhat smaller:

| | Hay 1958 | Hay 1959 | Hay 1960I | Hay 1960II |
|---------------------------|----------|----------|-----------|------------|
| before correction | 7 % | 1 % | 12 % | 5 % |
| after correction | 5 % | 1 % | 10 % | 4 % |

Obviously only a small part of the differences in $M_{m,500}$ between early and late hay may be explained by fermentation heat if this indeed always amounts to 80 % of the methane energy.

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SUMMARY AND CONCLUSIONS

CHAPTER I AND II. INTRODUCTION AND METHODS

The experiments were arranged according to the reversal method in 11 blocks of 4 single experiments each. In every block a comparison was made between early and late cut hay from the same pasture. In total four sets of early and late cut hay were examined with the aid of nine non-pregnant cows.

The accuracy per single experiment was calculated from within single experiment variation and from between single experiment variation (tables II 4-8). All standard deviations were small. The standard deviation calculated from within single experiment variation was the same or only a trifle smaller than that calculated from between single experiment variation as far as concerns digestibility, metabolisability and energy content of faeces, urine and methane. There was no evidence of between animal variation of digestibility and metabolisability. Especially in case of hay with low digestibility long experimental periods appear to be necessary if highest accuracy is to be obtained because variation increased with decreasing digestibility.

Variation of heat expenditure on successive days was clearly lower than with intervals of one week. There was little difference between the variations with intervals of one and of two weeks. The standard deviation of the heat expenditure per single experiment with six respiration days was estimated at 130 kcal or 1 % of the average daily heat production. This figure is calculated from within single experiment variation. Presumably between experiment variation yields a higher figure.

For the nitrogen balance the standard deviation per single experiment was also calculated from within single experiment variation. It amounted to ± 1.5 % of the average nitrogen intake. For the energy balance it was about 1.3 % of the gross energy intake.

The correlation between the energy balances computed a) from metabolisable energy, gaseous exchange and urine-N, and b) from N- and C-balance was very high. The latter balance usually was somewhat higher than the former.

CHAPTER III. EXPERIMENTAL

There were considerable differences between early and late cut hay in regard to composition and digestibility of dry matter and dry matter constituents, except for the hays of 1959; in this year there had been a severe drought. The range from lowest to highest digestibility was 53-77 for dry matter and 54-79 for organic matter. For all early and late cut hays the metabolisable energy was about 79 % of the digestible energy.

The loss of methane was about 8 % of the gross energy and 12.6 % of the digestible energy; it amounted to 5.2 g of CH₄ per 100 g of digestible carbohydrates (NFE + crude fibre). For the regression of methane energy (Y_2 , kcal) per 100 g of ingested dry matter on digestible dry matter (X_2 , g) per g of ingested dry matter we found

$$Y_2 = (32.3 \pm 4.4)X_2 + 13.4; \quad s_{Y_2X_2} = 1.0.$$

Several N-balances were slightly negative although for all hays except one the intake of digestible crude protein was considerably above the feeding standard of 70 g per 100 kg of body weight. In many cases this obviously was caused by shortage of energy intake resulting in negative energy balances.

25 out of 44 energy balances fluctuated between -1000 and +1000 kcal; only a few were lying below -2000 kcal or above +2000 kcal. With the aid of the results of preliminary digestion trials with sheep the range of the energy balances might be reduced.

CHAPTER IV. REGRESSION ANALYSIS

The data of the experiments were reduced to 500 kg live weight with KLEIBER's formula with power 3/4. Negative balances were multiplied by 0.83, because, according to MØLLGAARD, for prevention of negative gain 0.83 times as much metabolisable energy is needed as for positive gain.

In part of the calculations the energy balances have also been corrected to 12 hour standing (formula IV3) and (or) for so-called 'Produktionsausfall' (formula IV2). These two corrections proved to be immaterial.

Further reduction of the data was obtained by regression analysis with the aid of which the data were reduced to zero gain and average crude fibre intake (1717 g a day, corresponding to 33.55 % of crude fibre in the organic matter). The regression analysis was performed either by the ordinary methods or by VAN UVEN's method. With this method planes or hyperplanes of regression are calculated by minimising the sum of squares of the perpendiculars on these planes dropped from the points representing the observations.

The average maintenance requirement of an animal of 500 kg live weight proved to be 12000 kcal of metabolisable energy with standard deviation of 750 kcal. The large standard deviation is due to a combination of variations between animals and between periods. Especially variation between periods was highly significant.

Between period variations have also been recorded in the literature several times. They may be more systematic affecting all animals simultaneously. In other cases they do not go parallel: the level of maintenance may increase in one animal and decrease at the same time in another. Some possible causes of

these between period variations are mentioned. The necessity of studying these variations more closely in the near future is stressed.

Especially the systematic between period variations may be prejudicial to the reliability of the results of difference trials as in use for determining the fat production value of rations and single feeds. It is therefore recommended to eliminate in these difference trials not only between animal variation as is customary but also between period variation. For doing so the reversal methods (switch back methods) are recommended. The accuracy of the double reversal method is somewhat greater than that of the single reversal method covering only two periods, but in the former, work and cost are considerably greater.

As for the *maintenance requirement*, however, the reversal method ordinarily fails to increase the accuracy. Nevertheless for a problem like ours, i.e. the comparison of the *maintenance values* of early and late cut hay from the same field, this method proved to be very valuable.

The multiple regression of metabolisable energy on crude fibre (g) ingested and on gain (kcal) was studied. After elimination of between animal and between period variation we found with VAN UVEN's method of least squares and with zero gain a significant increase of 0.65 kcal of maintenance requirement per gram of crude fibre increase in the ration. The requirement of metabolisable energy per kcal of gain, with constant crude fibre intake of 1717 g a day, proved to be 1.49 kcal as calculated with the same method. This corresponds to an efficiency of 67.1%. With constant crude fibre *percentage* of 33.55% in the organic matter the efficiency of metabolisable energy for gain was found 60.5%.

Most of these values differ markedly from those found by KELLNER (1900), HOFFMANN et al. (1962) and BLAXTER (1961). A small part of the discrepancy could be explained by spurious correlation. After elimination of spurious correlation as good as possible VAN UVEN's method gave the following average regression formula (table IV 10):

$$M_{500} = \bar{M}_{m,500} + 1.5618G_{500} + 0.7160(F_{500} - 1717),$$

in which M_{500} = metabolisable energy (kcal) ingested,

$\bar{M}_{m,500}$ = metabolisable energy (kcal) requirement with zero gain and 1717 g of crude fibre in the ration,

F_{500} = crude fibre (g) in the ration.

All values refer to 500 kg body weight.

This formula shows an increase of maintenance requirement of 0.72 kcal per gram of crude fibre, thus somewhat more than without elimination of spurious correlation. However, even this figure is markedly lower than KELLNER's subtraction for crude fibre in lipogenesis amounting to about 2.2 kcal of meta-

bolisable energy per gram of crude fibre ingested. The whole body of evidence including our own experiments undoubtedly demonstrates that the requirement of metabolisable energy for maintenance is less dependent on crude fibre than requirement for fat synthesis. However, a certain degree of dependence cannot be denied completely.

After elimination of spurious correlation and with constant crude fibre intake of 1717 g a day the requirement of metabolisable energy per kcal of gain proved to be 1.56 kcal as calculated with VAN UVEN's formulae. This corresponds with an efficiency of 64.0 %, a figure differing not markedly from those found in difference trials with pure starch by KELLNER (1900) and by HOFFMANN et al. (1962) who found 62.6 % and 64.1 % respectively. The comparison with starch is reasonable because our figure 64.0 % refers to rations with constant crude fibre intake. Increase of metabolisable energy is therefore due to increased intake of crude fibre free organic matter, mainly starchy materials. It should however be mentioned that in the experiments of the authors quoted the basal rations consisted of roughage and concentrates whereas in our experiments they consisted of hay only.

With constant crude fibre *percentage* of 33.55 % in the organic matter the efficiency of metabolisable energy for gain proved to be 57.6 %, spurious correlation eliminated and calculated with VAN UVEN's method. This is still considerably higher than found by KELLNER for hay (41.5 and 46.2 %) and also higher than the value 38.1 % calculated for our hay with BLAXTER's formula (1961). Much better seems the agreement with the efficiency of 57.7 % found by NEHRING et al. (1961). However, in their experiments the crude fibre percentage of the organic matter was appreciably lower than in ours (25.33 versus 33.55 %).

There remains therefore a discrepancy, the reason of which is not clear to us. Possibly the efficiency of the Netherland hay for lipogenesis is higher than generally believed although presumably not as high as found by us up to now. It should however be kept in mind that highly accurate figures could not be expected because in planning our experiments it has been the intention to keep gain figures as small as possible in order to obtain more clear cut results for maintenance requirement. Obviously, more experiments with high hay rations will be necessary for clarification.

CHAPTER V. ANALYSIS WITH CORRECTION FACTORS

In chapter IV the efficiency of metabolisable energy for lipogenesis (energy gain) has been found rather high. In two difference trials (chapter V) with early and late cut hay 1959 the result was considerably lower: on an average 48 ± 5.5 % and 41 ± 6.9 % respectively. These figures being the result of only

two simultaneous difference trials are not sufficiently accurate for use as general correction factors for energy gain in all the experiments.

Therefore, in further calculations the metabolisable energy was corrected for energy gain with various efficiency factors from literature. Reductions for body weight to 500 kg was performed by multiplication by $(500/W)^p$. In total five combinations of efficiency factors and exponents p were used for computing the maintenance requirement of metabolisable energy per 500 kg live weight ($M_{m,500}$).

As there were 44 single respiration experiments, five sets of 44 maintenance values $M_{m,500}$ were obtained. By using these data for an analysis of variance significant variance of $M_{m,500}$ between early and late cut hay was found with the hays 1958, 1960I and 1960II; between animal plus between period variance was found significant with the hays 1960I and 1960II, almost regardless of the way in which $M_{m,500}$ had been computed (table V2). The use of the exponent 1 instead of 3/4 in the correction for body weight decreased all values $M_{m,500}$ by 2-4 %. A greater influence on the $M_{m,500}$ -values as well as on the differences of these values between early and late hay had the use of the various efficiency factors utilised in the correction for the energy balances (energy gain): the differences between the highest and lowest $M_{m,500}$ -values were 2-9 % of the average $M_{m,500}$. The differences between early and late cut hay were significant for the hays 1958, 1960I and 1960II for almost all efficiency factors; the higher $M_{m,500}$ -values belonged to the late hays. The average differences between early and late hay 1958, 1959, 1960I and 1960II were 7, 1, 12 and 5 % respectively. There was a high correlation between these differences and the differences of either the metabolisability ($r = -0.97$), the digestibility of dry matter and of energy ($r = -0.98$ and -0.97 respectively) or the crude fibre content ($r = +0.92$). The regressions of metabolisable energy requirement on crude fibre (grams ingested as well as percentage) were significant. The use of higher efficiency factors in correcting for energy gain coincided with lower regression of $M_{m,500}$ on crude fibre ingested.

The standard deviation of $M_{m,500}$ per single experiment amounted for the four hays to 4, 10, 6 and 3 % respectively when calculated from differences between early and late hay. Each of these last mentioned differences had been determined in one reversal trial as the difference of two successive single experiments with the same animal. If variation of heat expenditure was calculated from within single experiments the standard deviation of $M_{m,500}$ per single experiment was found somewhat lower. Presumably this is due to lower variance of heat production within single experiments than between single experiments.

If the assumption is correct that the rumen fermentation heat is always 80 % of the methane energy, this would explain the differences between the $M_{m,500}$ -values of early and late hay only for a small part.

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TABLE A. Data referring to individual cows on live weight, feed intake, digested feed constituents, digestion coefficients, urine, gaseous exchange, metabolisable energy and balances

TABLE A. Data referring to individual cows on live weight, feed intake, digested feed constituents, digestion coefficients, urine, gaseous exchange, metabolisable energy and balances

| No | Double period R | Sub-per. | Hay early cut (E) or late cut (L) | | Cow | Live weight (kg) | Intake (g or kcal) | | | | | | | No | Digested (g or kcal, %) | | | | | | | | | |
|-----|-----------------|----------|-----------------------------------|---|---------|------------------|--------------------|----------------------|-----------|-------------|--------------|-------------|------|-------|-------------------------|-------------|----------------|----------------------|------------|-------------|--------------|-------------|-------------|--------------|
| | | | | | | | dry matter | crude protein 6.25xN | crude fat | crude fibre | N-free extr. | min. matter | C | | kcal | dry matter | organic matter | crude protein 6.25xN | crude fat | crude fibre | N-free extr. | min. matter | C | kcal |
| | | | | | | | | | | | | | | | | | | | | | | | | |
| 1a | 17 | 1 | 1958 | E | Zwsch.1 | 598 | 6768 | 1114 | 210 | 1858 | 2767 | 819 | 2944 | 28823 | 1a | 4794 (70.8) | 4391 (73.8) | 772 (69.4) | 116 (55.1) | 1494 (80.4) | 2009 (72.6) | 403 (49.2) | 2068 (70.3) | 19907 (69.1) |
| 1b | 17 | 2 | 1958 | E | " | 598 | 6804 | 1120 | 211 | 1868 | 2782 | 823 | 2960 | 28978 | 1b | 4795 (70.5) | 4398 (73.5) | 778 (69.5) | 112 (53.2) | 1490 (79.8) | 2017 (72.5) | 397 (48.3) | 2069 (69.9) | 19902 (68.7) |
| 2a | 17 | 1 | 1958 | L | Zwk.2 | 549 | 7249 | 615 | 140 | 2747 | 3109 | 639 | 3243 | 31248 | 2a | 4360 (60.2) | 4085 (61.8) | 310 (50.4) | 58 (41.4) | 1892 (68.9) | 1824 (58.7) | 276 (43.2) | 1877 (57.9) | 17727 (56.7) |
| 2b | 17 | 2 | 1958 | L | " | 549 | 7573 | 644 | 145 | 2856 | 3236 | 694 | 3375 | 32503 | 2b | 4343 (57.3) | 4045 (58.8) | 302 (46.9) | 52 (35.9) | 1896 (66.4) | 1794 (55.4) | 299 (43.1) | 1844 (54.6) | 17343 (53.4) |
| 3a | 18 | 1 | 1958 | L | Zwsch.1 | 619 | 7941 | 677 | 155 | 2982 | 3406 | 721 | 3543 | 34116 | 3a | 4591 (57.8) | 4262 (59.0) | 325 (48.0) | 66 (42.4) | 1966 (65.9) | 1905 (55.9) | 330 (45.8) | 1956 (55.2) | 18432 (54.0) |
| 3b | 18 | 2 | 1958 | L | " | 619 | 7770 | 658 | 151 | 2910 | 3333 | 718 | 3458 | 33298 | 3b | 4603 (59.2) | 4254 (60.3) | 323 (49.1) | 64 (42.6) | 1946 (66.9) | 1921 (57.6) | 348 (48.5) | 1957 (56.6) | 18461 (55.4) |
| 4a | 18 | 1 | 1958 | E | Zwk.2 | 521 | 6298 | 1036 | 195 | 1729 | 2574 | 764 | 2739 | 26814 | 4a | 4578 (72.7) | 4192 (75.8) | 748 (72.2) | 113 (58.1) | 1414 (81.8) | 1917 (74.5) | 386 (50.5) | 1978 (72.2) | 19038 (71.0) |
| 4b | 18 | 2 | 1958 | E | " | 521 | 6298 | 1036 | 195 | 1729 | 2574 | 764 | 2739 | 26814 | 4b | 4559 (72.4) | 4183 (75.6) | 741 (71.5) | 113 (58.1) | 1412 (81.7) | 1918 (74.5) | 375 (49.1) | 1976 (72.1) | 18999 (70.9) |
| 5a | 19 | 1 | 1958 | L | Alke | 516 | 7229 | 606 | 131 | 2768 | 3050 | 673 | 3195 | 30815 | 5a | 4501 (62.3) | 4176 (63.7) | 304 (50.2) | 58 (44.4) | 1959 (70.8) | 1855 (60.8) | 324 (48.1) | 1935 (60.6) | 18312 (59.4) |
| 5b | 19 | 2 | 1958 | L | " | 516 | 6902 | 578 | 125 | 2637 | 2917 | 644 | 3054 | 29415 | 5b | 3829 (55.5) | 3598 (57.5) | 244 (42.3) | 42 (33.9) | 1737 (65.8) | 1574 (54.0) | 231 (35.9) | 1633 (53.5) | 15344 (52.2) |
| 6a | 19 | 1 | 1958 | E | Kee 2 | 540 | 6474 | 1064 | 191 | 1786 | 2635 | 798 | 2799 | 27452 | 6a | 4699 (72.6) | 4331 (76.3) | 756 (71.0) | 113 (59.3) | 1472 (82.4) | 1991 (75.5) | 368 (46.1) | 2047 (73.1) | 19783 (72.1) |
| 6b | 19 | 2 | 1958 | E | " | 540 | 6473 | 1064 | 191 | 1786 | 2635 | 798 | 2799 | 27448 | 6b | 4660 (72.0) | 4295 (75.7) | 751 (70.6) | 109 (57.3) | 1464 (82.0) | 1971 (74.8) | 365 (45.8) | 2023 (72.3) | 19558 (71.3) |
| 7a | 20 | 1 | 1958 | E | Alke | 488 | 6054 | 995 | 178 | 1670 | 2463 | 748 | 2617 | 25664 | 7a | 4313 (71.2) | 3982 (75.0) | 663 (66.6) | 91 (51.0) | 1355 (81.2) | 1873 (76.0) | 332 (44.3) | 1871 (71.5) | 18067 (70.4) |
| 7b | 20 | 2 | 1958 | E | " | 488 | 6048 | 994 | 178 | 1668 | 2461 | 747 | 2614 | 25636 | 7b | 4294 (71.0) | 3982 (75.1) | 662 (66.6) | 94 (52.6) | 1357 (81.4) | 1869 (76.0) | 312 (41.8) | 1874 (71.7) | 18094 (70.6) |
| 8a | 20 | 1 | 1958 | L | Kee 2 | 561 | 7072 | 594 | 130 | 2713 | 2992 | 643 | 3130 | 30168 | 8a | 4160 (58.8) | 3889 (60.5) | 280 (47.2) | 46 (35.1) | 1835 (67.6) | 1728 (57.8) | 271 (42.2) | 1774 (56.7) | 16642 (55.2) |
| 8b | 20 | 2 | 1958 | L | " | 561 | 6803 | 569 | 125 | 2604 | 2884 | 622 | 3012 | 29041 | 8b | 4101 (60.3) | 3826 (61.9) | 276 (48.6) | 49 (39.5) | 1790 (68.7) | 1711 (59.3) | 275 (44.2) | 1752 (58.2) | 16496 (56.8) |
| 9a | 25 | 1 | 1959 | E | Alke | 561 | 5793 | 930 | 187 | 1411 | 2699 | 567 | 2572 | 25261 | 9a | 4305 (74.3) | 4042 (77.4) | 647 (69.6) | 107 (57.5) | 1150 (81.6) | 2137 (79.2) | 263 (46.4) | 1908 (74.2) | 18411 (72.9) |
| 9b | 25 | 2 | 1959 | E | " | 561 | 5789 | 929 | 187 | 1410 | 2697 | 567 | 2571 | 25244 | 9b | 4186 (72.3) | 3951 (75.7) | 629 (67.7) | 99 (53.0) | 1128 (80.0) | 2095 (77.7) | 235 (41.5) | 1858 (72.3) | 17885 (70.8) |
| 10a | 25 | 1 | 1959 | L | Kee 2 | 611 | 6772 | 794 | 210 | 1857 | 3340 | 571 | 3002 | 29611 | 10a | 4748 (70.1) | 4489 (72.4) | 513 (64.6) | 121 (57.4) | 1326 (71.4) | 2529 (75.7) | 259 (45.5) | 2077 (69.2) | 20234 (68.3) |
| 10b | 25 | 2 | 1959 | L | " | 611 | 6764 | 793 | 210 | 1855 | 3336 | 570 | 2998 | 29578 | 10b | 4876 (72.1) | 4591 (74.1) | 529 (66.7) | 123 (58.6) | 1377 (74.2) | 2561 (76.8) | 286 (50.1) | 2140 (71.4) | 20793 (70.3) |
| 11a | 26 | 1 | 1959 | E | Alke | 591 | 8701 | 1380 | 292 | 2058 | 4117 | 853 | 3878 | 38124 | 11a | 6293 (72.3) | 5911 (75.3) | 937 (67.9) | 163 (55.9) | 1604 (77.9) | 3206 (77.9) | 383 (44.8) | 2796 (72.1) | 26914 (70.6) |
| 11b | 26 | 2 | 1959 | E | " | 591 | 8705 | 1380 | 292 | 2058 | 4117 | 858 | 3878 | 38121 | 11b | 6269 (72.0) | 5894 (75.1) | 913 (66.2) | 155 (53.2) | 1590 (77.3) | 3236 (78.6) | 375 (43.7) | 2778 (71.6) | 26789 (70.3) |
| 12a | 26 | 1 | 1959 | L | Kee 2 | 660 | 10016 | 1189 | 312 | 2741 | 5012 | 761 | 4519 | 44139 | 12a | 7018 (70.1) | 6687 (72.3) | 750 (63.1) | 180 (57.8) | 2002 (73.0) | 3754 (74.9) | 331 (43.5) | 3109 (68.8) | 29846 (67.6) |
| 12b | 26 | 2 | 1959 | L | " | 660 | 9989 | 1186 | 311 | 2735 | 5001 | 756 | 4509 | 44037 | 12b | 6972 (69.8) | 6645 (72.0) | 745 (62.8) | 169 (54.4) | 1954 (71.4) | 3776 (75.5) | 328 (43.4) | 3089 (68.5) | 29713 (67.5) |
| 13a | 27 | 1 | 1959 | L | Alke | 600 | 6684 | 794 | 201 | 1879 | 3267 | 542 | 3024 | 29451 | 13a | 4770 (71.4) | 4517 (73.5) | 506 (63.7) | 115 (57.3) | 1410 (75.0) | 2486 (76.1) | 254 (46.8) | 2123 (70.2) | 20310 (69.0) |
| 13b | 27 | 2 | 1959 | L | " | 600 | 6682 | 794 | 201 | 1879 | 3266 | 542 | 3023 | 29443 | 13b | 4663 (69.8) | 4425 (72.1) | 490 (61.7) | 133 (66.3) | 1374 (73.1) | 2428 (74.4) | 238 (43.8) | 2073 (68.6) | 19784 (67.2) |
| 14a | 27 | 1 | 1959 | E | Kee 2 | 619 | 6538 | 1054 | 258 | 1621 | 3007 | 598 | 2961 | 29058 | 14a | 4815 (73.6) | 4565 (76.9) | 729 (69.2) | 166 (64.6) | 1308 (80.7) | 2361 (78.5) | 250 (41.8) | 2181 (73.7) | 21042 (72.4) |
| 14b | 27 | 2 | 1959 | E | " | 619 | 6540 | 1054 | 258 | 1621 | 3007 | 600 | 2962 | 29059 | 14b | 4737 (72.4) | 4492 (75.6) | 725 (68.8) | 185 (71.8) | 1279 (78.9) | 2304 (76.6) | 245 (40.8) | 2142 (72.3) | 20648 (71.1) |
| 15a | 29 | 1 | 1959 | E | RW 12 | 566 | 5393 | 854 | 163 | 1310 | 2553 | 513 | 2429 | 23879 | 15a | 3994 (74.1) | 3755 (76.9) | 574 (67.3) | 93 (56.8) | 1052 (80.3) | 2036 (79.7) | 239 (46.6) | 1793 (73.8) | 17312 (72.5) |
| 15b | 29 | 2 | 1959 | E | " | 566 | 5390 | 853 | 163 | 1310 | 2552 | 512 | 2428 | 23866 | 15b | 3837 (71.2) | 3636 (74.5) | 553 (64.8) | 84 (51.6) | 1015 (77.5) | 1984 (77.7) | 202 (39.4) | 1724 (71.0) | 16647 (69.8) |
| 16a | 29 | 1 | 1959 | L | Zwsch.2 | 575 | 5908 | 695 | 162 | 1630 | 2934 | 487 | 2675 | 26015 | 16a | 4090 (69.2) | 3884 (71.6) | 421 (60.6) | 87 (53.5) | 1182 (72.6) | 2194 (74.8) | 206 (42.3) | 1826 (68.3) | 17382 (66.8) |
| 16b | 29 | 2 | 1959 | L | " | 575 | 5909 | 695 | 162 | 1630 | 2935 | 488 | 2675 | 26019 | 16b | 4144 (70.1) | 3926 (72.4) | 431 (62.1) | 87 (53.9) | 1180 (72.4) | 2227 (75.9) | 218 (44.7) | 1845 (69.0) | 17620 (67.7) |
| 17a | 30 | 1 | 1959 | L | RW 12 | 567 | 5908 | 689 | 165 | 1623 | 2934 | 497 | 2643 | 25917 | 17a | 4202 (71.1) | 3958 (73.1) | 437 (63.4) | 94 (57.4) | 1181 (72.8) | 2245 (76.5) | 244 (49.1) | 1843 (69.7) | 17802 (68.7) |
| 17b | 30 | 2 | 1959 | L | " | 567 | 5909 | 689 | 165 | 1623 | 2935 | 497 | 2643 | 25922 | 17b | 4027 (68.2) | 3812 (70.4) | 408 (59.3) | 88 (53.3) | 1123 (69.2) | 2192 (74.7) | 216 (43.3) | 1767 (66.8) | 17003 (65.6) |
| 18a | 30 | 1 | 1959 | E | Zwsch.2 | 562 | 5435 | 857 | 162 | 1311 | 2562 | 542 | 2430 | 23885 | 18a | 4064 (74.8) | 3812 (77.9) | 599 (69.9) | 95 (58.6) | 1082 (82.5) | 2036 (79.5) | 252 (46.5) | 1818 (74.8) | 17594 (73.7) |
| 18b | 30 | 2 | 1959 | E | " | 562 | 5433 | 857 | 162 | 1311 | 2561 | 542 | 2429 | 23877 | 18b | 3959 (72.9) | 3707 (75.8) | 578 (67.4) | 88 (54.6) | 1048 (80.0) | 1992 (77.8) | 252 (46.5) | 1757 (72.3) | 16994 (71.2) |

TABLE A. Continued

| No | Double period R | Sub-per. | Hay early cut (E) or late cut (L) | | Cow | Live weight (kg) | Intake (g or kcal) | | | | | | | No | Digested (g or kcal, %) | | | | | | | | | |
|-----|-----------------|----------|--|---|---------|------------------|--------------------|----------------------|-----------|-------------|--------------|-------------|------|-------|-------------------------|-------------|----------------|----------------------|------------|-------------|--------------|-------------|-------------|--------------|
| | | | | | | | dry matter | crude protein 6.25xN | crude fat | crude fibre | N-free extr. | min. matter | C | | kcal | dry matter | organic matter | crude protein 6.25xN | crude fat | crude fibre | N-free extr. | min. matter | C | kcal |
| | | | | | | | | | | | | | | | | | | | | | | | | |
| 19a | 31 | 1 | 1958 | E | RW 12. | 562 | 5790 | 958 | 164 | 1639 | 2298 | 730 | 2522 | 24785 | 19a | 4058 (70.1) | 3715 (73.4) | 625 (65.2) | 85 (51.6) | 1326 (80.9) | 1679 (73.1) | 343 (46.9) | 1769 (70.1) | 17083 (68.9) |
| 19b | 31 | 2 | 1958 | E | " | 562 | 5790 | 958 | 164 | 1639 | 2298 | 730 | 2522 | 24785 | 19b | 4056 (70.1) | 3726 (73.6) | 624 (65.2) | 86 (52.7) | 1344 (82.0) | 1672 (72.7) | 330 (45.1) | 1772 (70.3) | 17141 (69.2) |
| 20a | 31 | 1 | 1958 | L | Zwsch.2 | 585 | 8260 | 701 | 142 | 3228 | 3450 | 739 | 3714 | 35739 | 20a | 4861 (58.9) | 4533 (60.3) | 321 (45.8) | 52 (36.9) | 2210 (68.5) | 1950 (56.5) | 328 (44.4) | 2108 (56.7) | 19842 (55.5) |
| 20b | 31 | 2 | 1958 | L | " | 585 | 8252 | 702 | 142 | 3231 | 3453 | 724 | 3718 | 35773 | 20b | 4721 (57.2) | 4427 (58.8) | 301 (42.9) | 54 (37.9) | 2167 (67.1) | 1905 (55.2) | 293 (40.5) | 2046 (55.0) | 19297 (53.9) |
| 21a | 32 | 1 | 1958 | L | RW 12 | 591 | 8497 | 718 | 146 | 3307 | 3534 | 792 | 3805 | 36617 | 21a | 4825 (56.8) | 4491 (58.3) | 330 (45.9) | 49 (33.9) | 2158 (65.2) | 1954 (55.3) | 334 (42.2) | 2092 (55.0) | 19657 (53.7) |
| 21b | 32 | 2 | 1958 | L | " | 591 | 8462 | 715 | 145 | 3295 | 3521 | 786 | 3791 | 36476 | 21b | 4803 (56.8) | 4473 (58.3) | 324 (45.4) | 44 (30.7) | 2174 (66.0) | 1930 (54.8) | 330 (41.9) | 2078 (54.8) | 19587 (53.7) |
| 22a | 32 | 1 | 1958 | E | Zwsch.2 | 559 | 5791 | 958 | 164 | 1640 | 2299 | 730 | 2522 | 24790 | 22a | 4019 (69.4) | 3716 (73.4) | 624 (65.1) | 78 (47.2) | 1336 (81.5) | 1678 (73.0) | 303 (41.5) | 1759 (69.7) | 17010 (68.6) |
| 22b | 32 | 2 | 1958 | E | " | 559 | 5788 | 958 | 164 | 1639 | 2298 | 730 | 2521 | 24778 | 22b | 4141 (71.5) | 3809 (75.3) | 643 (67.2) | 86 (52.5) | 1363 (83.2) | 1716 (74.7) | 332 (45.5) | 1816 (72.0) | 17584 (71.0) |
| 23 | 33 | | 1960I | E | Kee 3 | 529 | 5073 | 1004 | 193 | 1083 | 2312 | 482 | 2304 | 22849 | 23 | 3867 (76.2) | 3614 (78.7) | 733 (73.1) | 108 (55.9) | 886 (81.8) | 1887 (81.6) | 253 (52.5) | 1742 (75.6) | 17032 (74.5) |
| 24 | 33 | | 1960I | E | Alke | 604 | 5064 | 1002 | 192 | 1082 | 2308 | 480 | 2302 | 22818 | 24 | 3872 (76.5) | 3621 (79.0) | 732 (73.0) | 108 (56.2) | 881 (81.5) | 1899 (82.3) | 252 (52.5) | 1742 (75.7) | 16971 (74.4) |
| 25 | 33 | | 1960I | L | RW 12 | 576 | 6365 | 743 | 144 | 2174 | 2744 | 560 | 2862 | 27846 | 25 | 3707 (58.2) | 3496 (60.2) | 432 (58.1) | 69 (47.7) | 1383 (63.6) | 1612 (58.8) | 211 (37.7) | 1615 (56.4) | 15393 (55.3) |
| 26 | 33 | | 1960I | L | Zwsch.2 | 585 | 6347 | 741 | 144 | 2170 | 2738 | 553 | 2857 | 27794 | 26 | 3838 (60.5) | 3610 (62.3) | 440 (59.3) | 70 (48.9) | 1420 (65.4) | 1680 (61.3) | 228 (41.3) | 1691 (59.2) | 16158 (58.1) |
| 27 | 34 | | 1960I | L | Kee 3 | 561 | 6420 | 744 | 129 | 2185 | 2782 | 579 | 2882 | 28018 | 27 | 3804 (59.2) | 3559 (60.9) | 434 (58.4) | 51 (39.7) | 1422 (65.1) | 1651 (59.3) | 245 (42.3) | 1654 (57.4) | 15793 (56.4) |
| 28 | 34 | | 1960I | L | Alke | 613 | 6436 | 746 | 130 | 2188 | 2786 | 586 | 2887 | 28062 | 28 | 3806 (59.1) | 3587 (61.3) | 431 (57.8) | 50 (38.7) | 1401 (64.0) | 1706 (61.2) | 219 (37.4) | 1669 (57.8) | 15956 (56.9) |
| 29 | 34 | | 1960I | E | RW 12 | 566 | 5102 | 1016 | 191 | 1108 | 2301 | 486 | 2316 | 22965 | 29 | 3934 (77.1) | 3678 (79.7) | 756 (74.5) | 109 (56.8) | 933 (84.2) | 1880 (81.7) | 256 (52.6) | 1771 (76.5) | 17295 (75.3) |
| 30 | 34 | | 1960I | E | Zwsch.2 | 551 | 5107 | 1016 | 192 | 1109 | 2303 | 487 | 2318 | 22986 | 30 | 3955 (77.4) | 3701 (80.1) | 769 (75.7) | 113 (59.1) | 926 (83.5) | 1892 (82.2) | 254 (52.2) | 1781 (76.8) | 17389 (75.7) |
| 31 | 35 | | 1960II | E | Kee 3 | 541 | 5572 | 1040 | 134 | 1646 | 2164 | 588 | 2486 | 24713 | 31 | 3489 (62.6) | 3217 (64.6) | 689 (66.3) | 48 (35.5) | 1198 (72.8) | 1281 (59.2) | 273 (46.3) | 1506 (60.6) | 14776 (59.8) |
| 32 | 35 | | 1960II | E | Alke | 597 | 5567 | 1038 | 134 | 1644 | 2162 | 588 | 2484 | 24691 | 32 | 3516 (63.2) | 3245 (65.2) | 692 (66.7) | 48 (35.8) | 1181 (71.8) | 1324 (61.3) | 271 (46.0) | 1521 (61.2) | 14870 (60.2) |
| 33 | 35 | | 1960II | L | RW 12 | 571 | 6373 | 873 | 128 | 2147 | 2596 | 630 | 2832 | 27849 | 33 | 3452 (54.2) | 3193 (55.6) | 499 (57.2) | 34 (27.0) | 1296 (60.3) | 1364 (52.6) | 259 (41.1) | 1468 (51.8) | 14276 (51.3) |
| 34 | 35 | | 1960II | L | Zwsch.2 | 584 | 6333 | 868 | 127 | 2136 | 2583 | 620 | 2818 | 27705 | 34 | 3447 (54.4) | 3201 (56.0) | 498 (57.3) | 40 (31.8) | 1302 (61.0) | 1361 (52.7) | 246 (39.7) | 1467 (52.1) | 14243 (51.4) |
| 35 | 36 | | 1960II | L | Kee 3 | 548 | 6325 | 872 | 132 | 2153 | 2559 | 609 | 2836 | 27672 | 35 | 3359 (53.1) | 3116 (54.5) | 488 (55.9) | 46 (35.2) | 1291 (59.9) | 1291 (50.5) | 243 (40.0) | 1447 (51.0) | 13862 (50.1) |
| 36 | 36 | | 1960II | L | Alke | 602 | 6516 | 895 | 135 | 2209 | 2625 | 652 | 2909 | 28390 | 36 | 3484 (53.5) | 3224 (55.0) | 501 (56.0) | 47 (35.0) | 1375 (62.3) | 1300 (49.5) | 260 (39.9) | 1492 (51.3) | 14299 (50.4) |
| 37 | 36 | | 1960II | E | RW 12 | 566 | 5681 | 1057 | 143 | 1632 | 2250 | 599 | 2539 | 25160 | 37 | 3646 (64.2) | 3389 (66.7) | 696 (65.8) | 54 (37.6) | 1170 (71.7) | 1469 (65.3) | 257 (43.0) | 1580 (62.2) | 15431 (61.3) |
| 38 | 36 | | 1960II | E | Zwsch.2 | 558 | 5691 | 1059 | 143 | 1634 | 2253 | 602 | 2542 | 25197 | 38 | 3686 (64.8) | 3396 (66.7) | 717 (67.7) | 55 (38.1) | 1196 (73.2) | 1428 (63.4) | 290 (48.2) | 1596 (62.8) | 15572 (61.8) |
| 39 | 38 | | 1960I | E | Jansje | 566 | 4926 | 970 | 175 | 1044 | 2268 | 469 | 2225 | 22132 | 39 | 3757 (76.3) | 3498 (78.5) | 694 (71.5) | 97 (55.2) | 868 (83.1) | 1840 (81.1) | 259 (55.3) | 1673 (75.2) | 16407 (74.1) |
| 40 | 38 | | 1960I | L | Roosje | 594 | 6731 | 769 | 137 | 2184 | 3093 | 548 | 3031 | 29555 | 40 | 3917 (58.2) | 3715 (60.1) | 434 (56.4) | 53 (38.7) | 1421 (65.1) | 1807 (58.4) | 202 (36.9) | 1716 (56.6) | 16428 (55.6) |
| 41 | 39 | | 1960I | L | Jansje | 587 | 6376 | 730 | 125 | 2133 | 2911 | 476 | 2891 | 28080 | 41 | 3746 (58.8) | 3555 (60.3) | 406 (55.6) | 50 (39.8) | 1408 (66.0) | 1692 (58.1) | 191 (40.1) | 1634 (56.5) | 15586 (55.5) |
| 42 | 39 | | 1960I | E | Roosje | 545 | 4969 | 973 | 173 | 1050 | 2303 | 471 | 2232 | 22229 | 42 | 3802 (76.5) | 3541 (78.7) | 720 (74.0) | 100 (57.6) | 884 (84.2) | 1837 (79.8) | 261 (55.5) | 1680 (75.3) | 16481 (74.1) |
| 43 | 40 | | 1960II | E | Jansje | 568 | 6139 | 1153 | 149 | 1718 | 2480 | 640 | 2728 | 27119 | 43 | 3944 (64.2) | 3621 (65.8) | 761 (66.0) | 58 (39.0) | 1276 (74.3) | 1525 (61.5) | 324 (50.6) | 1690 (61.9) | 16559 (61.1) |
| 44 | 40 | | 1960II | L | Roosje | 573 | 7389 | 1030 | 146 | 2372 | 3097 | 744 | 3285 | 32155 | 44 | 3776 (51.1) | 3492 (52.5) | 561 (54.4) | 50 (34.5) | 1418 (59.8) | 1463 (47.2) | 284 (38.2) | 1613 (49.1) | 15410 (47.9) |
| 45 | 41 | | 1960II | L | Jansje | 572 | 6796 | 917 | 128 | 2200 | 2898 | 652 | 3058 | 29706 | 45 | 3558 (52.4) | 3294 (53.6) | 472 (51.5) | 35 (27.2) | 1327 (60.3) | 1460 (50.4) | 264 (40.5) | 1525 (49.9) | 14624 (49.2) |
| 46 | 41 | | 1960II | E | Roosje | 541 | 6080 | 1128 | 143 | 1716 | 2462 | 631 | 2737 | 26889 | 46 | 3824 (62.9) | 3526 (64.7) | 741 (65.7) | 53 (37.2) | 1261 (73.5) | 1470 (59.7) | 298 (47.2) | 1676 (61.3) | 16097 (59.9) |

TABLE A. Continued

| No | Double period R | Sub-per. | Hay early cut (E) or late cut (L) | | Cow | Live weight (kg) | Urine | | | | Gaseous exchange (CO ₂ in urine included) | | | | Metab. energy | | Balance (g or kcal) | | | | |
|-----|-----------------|----------|--|---|---------|------------------|--------|-----|------|-----------------|---|-----------------|-----------------|------|---------------|-------|---------------------|--------|--------|--------|----------|
| | | | | | | | 6.25xN | C | kcal | CO ₂ | O ₂ | CO ₂ | CH ₄ | | | | 6.25xN | C | energy | | |
| | | | | | | | | | | | | | ltr | ltr | | | | | ltr | kcal | kcal (%) |
| 1a | 17 | 1 | 1958 | E | Zwsch.1 | 598 | 766 | 186 | 1855 | 43 | 2930 | 3134 | 261 | 2466 | 15586 (54.1) | + 6 | + 85 | + 1053 | + 811 | + 932 | |
| 1b | 17 | 2 | 1958 | E | " | 598 | 775 | 188 | 1885 | 52 | 2910 | 3155 | 256 | 2419 | 15598 (53.8) | + 3 | + 80 | + 987 | + 879 | + 933 | |
| 2a | 17 | 1 | 1958 | L | Zwk.2 | 549 | 384 | 120 | 1191 | 22 | 2699 | 2956 | 284 | 2684 | 13852 (44.3) | - 74 | + 32 | + 446 | + 111 | + 278 | |
| 2b | 17 | 2 | 1958 | L | " | 549 | 367 | 120 | 1086 | 32 | 2794 | 3086 | 293 | 2769 | 13488 (41.5) | - 65 | - 71 | - 828 | - 776 | - 802 | |
| 3a | 18 | 1 | 1958 | L | Zwsch.1 | 619 | 361 | 124 | 1150 | 24 | 2990 | 3166 | 274 | 2589 | 14693 (43.1) | - 36 | + 1 | + 36 | - 437 | - 200 | |
| 3b | 18 | 2 | 1958 | L | " | 619 | 335 | 120 | 1091 | 25 | 2883 | 3100 | 276 | 2613 | 14757 (44.3) | - 12 | + 39 | + 495 | + 116 | + 306 | |
| 4a | 18 | 1 | 1958 | E | Zwk.2 | 521 | 752 | 178 | 1776 | 48 | 2708 | 2938 | 250 | 2367 | 14896 (55.6) | - 5 | + 116 | + 1438 | + 1207 | + 1322 | |
| 4b | 18 | 2 | 1958 | E | " | 521 | 742 | 176 | 1765 | 72 | 2680 | 2942 | 248 | 2348 | 14886 (55.5) | - 1 | + 128 | + 1580 | + 1299 | + 1440 | |
| 5a | 19 | 1 | 1958 | L | Alke | 516 | 345 | 116 | 1082 | 19 | 2712 | 2917 | 258 | 2433 | 14797 (48.0) | - 41 | + 128 | + 1608 | + 1031 | + 1320 | |
| 5b | 19 | 2 | 1958 | L | " | 516 | 372 | 116 | 1097 | 24 | 2736 | 2978 | 264 | 2490 | 11757 (40.0) | - 127 | - 209 | - 2490 | - 2165 | - 2327 | |
| 6a | 19 | 1 | 1958 | E | Kee 2 | 540 | 802 | 184 | 1709 | 37 | 2757 | 2996 | 250 | 2358 | 15716 (57.2) | - 47 | + 142 | + 1790 | + 1779 | + 1784 | |
| 6b | 19 | 2 | 1958 | E | " | 540 | 783 | 183 | 1804 | 42 | 2749 | 3004 | 258 | 2438 | 15316 (55.8) | - 32 | + 113 | + 1428 | + 1402 | + 1415 | |
| 7a | 20 | 1 | 1958 | E | Alke | 488 | 685 | 164 | 1599 | 33 | 2566 | 2761 | 220 | 2074 | 14394 (56.1) | - 22 | + 127 | + 1589 | + 1438 | + 1514 | |
| 7b | 20 | 2 | 1958 | E | " | 488 | 624 | 156 | 1501 | 37 | 2586 | 2792 | 228 | 2150 | 14443 (56.3) | + 38 | + 118 | + 1439 | + 1361 | + 1400 | |
| 8a | 20 | 1 | 1958 | L | Kee 2 | 561 | 354 | 117 | 1144 | 16 | 2707 | 2933 | 262 | 2471 | 13026 (43.2) | - 74 | - 47 | - 526 | - 737 | - 631 | |
| 8b | 20 | 2 | 1958 | L | " | 561 | 365 | 113 | 1070 | 16 | 2547 | 2696 | 234 | 2216 | 13211 (45.5) | - 89 | + 76 | + 1009 | + 339 | + 674 | |
| 9a | 25 | 1 | 1959 | E | Alke | 561 | 620 | 172 | 1650 | 41 | 2420 | 2669 | 223 | 2104 | 14657 (58.0) | + 27 | + 208 | + 2551 | + 2359 | + 2455 | |
| 9b | 25 | 2 | 1959 | E | " | 561 | 620 | 178 | 1647 | 43 | 2462 | 2719 | 237 | 2240 | 13999 (55.5) | + 9 | + 118 | + 1458 | + 1489 | + 1474 | |
| 10a | 25 | 1 | 1959 | L | Kee 2 | 611 | 507 | 173 | 1630 | 27 | 2632 | 2958 | 255 | 2407 | 16198 (54.7) | + 6 | + 195 | + 2412 | + 2727 | + 2569 | |
| 10b | 25 | 2 | 1959 | L | " | 611 | 482 | 173 | 1496 | 30 | 2625 | 2961 | 254 | 2400 | 16897 (57.1) | + 47 | + 260 | + 3180 | + 3443 | + 3312 | |
| 11a | 26 | 1 | 1959 | E | Alke | 591 | 827 | 240 | 2290 | 58 | 3212 | 3673 | 310 | 2932 | 21692 (56.9) | + 110 | + 451 | + 5500 | + 5225 | + 5363 | |
| 11b | 26 | 2 | 1959 | E | " | 591 | 859 | 246 | 2256 | 77 | 3248 | 3718 | 306 | 2889 | 21644 (56.8) | + 54 | + 415 | + 5101 | + 4986 | + 5043 | |
| 12a | 26 | 1 | 1959 | L | Kee 2 | 660 | 702 | 252 | 2412 | 46 | 3524 | 4159 | 378 | 3575 | 23859 (54.1) | + 49 | + 449 | + 5517 | + 5610 | + 5563 | |
| 12b | 26 | 2 | 1959 | L | " | 660 | 721 | 242 | 2342 | 47 | 3553 | 4167 | 371 | 3503 | 23868 (54.2) | + 24 | + 440 | + 5424 | + 5498 | + 5461 | |
| 13a | 27 | 1 | 1959 | L | Alke | 600 | 500 | 171 | 1650 | 44 | 2781 | 3069 | 265 | 2507 | 16153 (54.8) | + 6 | + 188 | + 2319 | + 1976 | + 2148 | |
| 13b | 27 | 2 | 1959 | L | " | 600 | 465 | 168 | 1588 | 42 | 2754 | 3040 | 260 | 2454 | 15742 (53.5) | + 25 | + 159 | + 1945 | + 1693 | + 1819 | |
| 14a | 27 | 1 | 1959 | E | Kee 2 | 619 | 709 | 203 | 1957 | 70 | 2938 | 3218 | 256 | 2419 | 16665 (57.4) | + 21 | + 153 | + 1880 | + 1744 | + 1812 | |
| 14b | 27 | 2 | 1959 | E | " | 619 | 720 | 199 | 1922 | 36 | 2909 | 3129 | 248 | 2346 | 16379 (56.4) | + 4 | + 152 | + 1873 | + 1676 | + 1774 | |
| 15a | 29 | 1 | 1959 | E | RW 12 | 566 | 586 | 165 | 1573 | 40 | 2606 | 2737 | 203 | 1916 | 13824 (57.9) | - 11 | + 73 | + 911 | + 709 | + 810 | |
| 15b | 29 | 2 | 1959 | E | " | 566 | 586 | 167 | 1583 | 60 | 2684 | 2788 | 207 | 1959 | 13105 (54.9) | - 33 | - 17 | - 190 | - 370 | - 280 | |
| 16a | 29 | 1 | 1959 | L | Zwsch.2 | 575 | 473 | 160 | 1502 | 45 | 2819 | 2975 | 231 | 2186 | 13694 (52.6) | - 52 | - 29 | - 316 | - 541 | - 429 | |
| 16b | 29 | 2 | 1959 | L | " | 575 | 480 | 159 | 1521 | 48 | 2772 | 2936 | 240 | 2265 | 13834 (53.2) | - 49 | + 9 | + 149 | - 166 | - 8 | |
| 17a | 30 | 1 | 1959 | L | RW 12 | 567 | 406 | 151 | 1394 | 32 | 2665 | 2811 | 222 | 2098 | 14310 (55.2) | + 31 | + 83 | + 1002 | + 847 | + 925 | |
| 17b | 30 | 2 | 1959 | L | " | 567 | 432 | 149 | 1380 | 47 | 2681 | 2853 | 219 | 2067 | 13556 (52.3) | - 24 | - 4 | - 32 | - 16 | - 24 | |
| 18a | 30 | 1 | 1959 | E | Zwsch.2 | 562 | 612 | 167 | 1574 | 55 | 2617 | 2735 | 213 | 2013 | 14008 (58.6) | - 13 | + 100 | + 1243 | + 862 | + 1052 | |
| 18b | 30 | 2 | 1959 | E | " | 562 | 580 | 171 | 1528 | 62 | 2568 | 2731 | 215 | 2035 | 13431 (56.3) | - 2 | + 39 | + 488 | + 474 | + 481 | |

TABLE A. Continued

| No | Double period R | Sub-per. | Hay early cut(E) or late cut(L) | | Cow | Live weight (kg) | Urine | | | | Gaseous exchange (CO ₂ in urine included) | | | | Metab. energy kcal (%) | Balance (g or kcal) | | | | |
|-----|-----------------|----------|--|---|---------|------------------|-------------|--------|------|------------------------|---|------------------------|-----------------|------|---------------------------|---------------------|-------|--------------|----------------|---------|
| | | | | | | | 6.25xN g | C g | kcal | CO ₂ ltr | O ₂ ltr | CO ₂ ltr | CH ₄ | | | 6.25xN | C | energy | | |
| | | | | | | | | | | | | | ltr | kcal | | | | from CN-bal. | from heat det. | average |
| 19a | 31 | 1 | 1958 | E | RW 12 | 562 | 612 | 153 | 1483 | 57 | 2667 | 2807 | 215 | 2032 | 13568 (54.7) | + 13 | + 26 | + 306 | + 146 | + 226 |
| 19b | 31 | 2 | 1958 | E | " | 562 | 620 | 154 | 1462 | 60 | 2586 | 2758 | 219 | 2070 | 13610 (54.9) | + 5 | + 54 | + 670 | + 562 | + 616 |
| 20a | 31 | 1 | 1958 | L | Zwsch.2 | 585 | 288 | 118 | 1070 | 32 | 2889 | 3157 | 282 | 2668 | 16104 (45.1) | + 33 | + 163 | + 1988 | + 1365 | + 1677 |
| 20b | 31 | 2 | 1958 | L | " | 585 | 294 | 118 | 1015 | 32 | 2941 | 3174 | 278 | 2627 | 15655 (43.8) | + 7 | + 94 | + 1157 | + 694 | + 926 |
| 21a | 32 | 1 | 1958 | L | RW 12 | 591 | 324 | 123 | 1105 | 20 | 2970 | 3230 | 285 | 2696 | 15856 (43.3) | + 6 | + 94 | + 1161 | + 726 | + 944 |
| 21b | 32 | 2 | 1958 | L | " | 591 | 339 | 124 | 1115 | 33 | 2968 | 3206 | 285 | 2690 | 15782 (43.3) | - 15 | + 100 | + 1251 | + 691 | + 971 |
| 22a | 32 | 1 | 1958 | E | Zwsch.2 | 559 | 664 | 159 | 1556 | 45 | 2527 | 2709 | 227 | 2142 | 13311 (53.7) | - 40 | + 50 | + 642 | + 565 | + 604 |
| 22b | 32 | 2 | 1958 | E | " | 559 | 635 | 158 | 1492 | 70 | 2536 | 2710 | 215 | 2035 | 14056 (56.7) | + 8 | + 127 | + 1563 | + 1262 | + 1413 |
| 23 | 33 | | 1960I | E | Kee 3 | 529 | 731 | 181 | 1558 | 67 | 2545 | 2632 | 192 | 1810 | 13664 (59.8) | + 2 | + 83 | + 1021 | + 936 | + 978 |
| 24 | 33 | | 1960I | E | Alke | 604 | 720 | 147 | 1583 | 54 | 2532 | 2624 | 201 | 1897 | 13492 (59.1) | + 12 | + 108 | + 1332 | + 826 | + 1079 |
| 25 | 33 | | 1960I | L | RW 12 | 576 | 481 | 124 | 1151 | 41 | 2554 | 2678 | 210 | 1987 | 12255 (44.0) | - 50 | - 35 | - 398 | - 606 | - 502 |
| 26 | 33 | | 1960I | L | Zwsch.2 | 585 | 492 | 130 | 1147 | 45 | 2525 | 2680 | 213 | 2011 | 13000 (46.8) | - 52 | + 34 | + 458 | + 249 | + 353 |
| 27 | 34 | | 1960I | L | Kee 3 | 561 | 458 | 125 | 1140 | 39 | 2492 | 2651 | 215 | 2030 | 12624 (45.1) | - 24 | + 13 | + 181 | + 28 | + 104 |
| 28 | 34 | | 1960I | L | Alke | 613 | 454 | 125 | 1191 | 38 | 2599 | 2715 | 220 | 2076 | 12690 (45.2) | - 24 | - 9 | - 96 | - 393 | - 245 |
| 29 | 34 | | 1960I | E | RW 12 | 566 | 711 | 172 | 1680 | 66 | 2362 | 2531 | 200 | 1890 | 13725 (59.8) | + 45 | + 171 | + 2084 | + 1825 | + 1954 |
| 30 | 34 | | 1960I | E | Zwsch.2 | 551 | 748 | 178 | 1750 | 65 | 2348 | 2525 | 204 | 1926 | 13714 (59.7) | + 21 | + 175 | + 2147 | + 1885 | + 2016 |
| 31 | 35 | | 1960II | E | Kee 3 | 541 | 698 | 145 | 1438 | 34 | 2362 | 2439 | 189 | 1784 | 11554 (46.8) | - 8 | - 29 | - 355 | - 243 | - 299 |
| 32 | 35 | | 1960II | E | Alke | 597 | 735 | 149 | 1564 | 17 | 2392 | 2472 | 198 | 1875 | 11431 (46.3) | - 43 | - 51 | - 595 | - 509 | - 552 |
| 33 | 35 | | 1960II | L | RW 12 | 571 | 527 | 111 | 1140 | 14 | 2381 | 2459 | 198 | 1876 | 11260 (40.4) | - 28 | - 61 | - 730 | - 669 | - 700 |
| 34 | 35 | | 1960II | L | Zwsch.2 | 584 | 552 | 114 | 1158 | 20 | 2384 | 2468 | 204 | 1923 | 11162 (40.3) | - 55 | - 69 | - 816 | - 779 | - 798 |
| 35 | 36 | | 1960II | L | Kee 3 | 548 | 513 | 110 | 1134 | 11 | 2391 | 2456 | 194 | 1835 | 10893 (39.4) | - 26 | - 78 | - 952 | - 1078 | - 1015 |
| 36 | 36 | | 1960II | L | Alke | 602 | 535 | 116 | 1205 | 17 | 2543 | 2600 | 201 | 1901 | 11192 (39.4) | - 34 | - 117 | - 1427 | - 1529 | - 1478 |
| 37 | 36 | | 1960II | E | RW 12 | 566 | 694 | 146 | 1521 | 28 | 2392 | 2512 | 202 | 1914 | 11996 (47.7) | + 1 | - 6 | - 75 | + 1 | - 37 |
| 38 | 36 | | 1960II | E | Zwsch.2 | 558 | 752 | 153 | 1584 | 32 | 2414 | 2522 | 204 | 1925 | 12063 (47.9) | - 35 | - 2 | - 1 | - 14 | - 8 |
| 39 | 38 | | 1960I | E | Jansje | 566 | 649 | 164 | 1589 | 39 | 2716 | 2766 | 200 | 1885 | 12933 (58.4) | + 45 | - 60 | - 770 | - 631 | - 700 |
| 40 | 38 | | 1960I | L | Roosje | 594 | 533 | 140 | 1310 | 33 | 2981 | 3056 | 234 | 2207 | 12912 (43.7) | - 99 | - 170 | - 2035 | - 2031 | - 2033 |
| 41 | 39 | | 1960I | L | Jansje | 587 | 420 | 122 | 1139 | 17 | 2778 | 2850 | 228 | 2155 | 12292 (43.8) | - 14 | - 129 | - 1592 | - 1648 | - 1620 |
| 42 | 39 | | 1960I | E | Roosje | 545 | 748 | 176 | 1698 | 44 | 2639 | 2696 | 202 | 1909 | 12874 (57.9) | - 28 | - 27 | - 310 | - 286 | - 298 |
| 43 | 40 | | 1960II | E | Jansje | 568 | 741 | 155 | 1628 | 22 | 2805 | 2812 | 212 | 2002 | 12929 (47.7) | + 19 | - 75 | - 944 | - 1008 | - 976 |
| 44 | 40 | | 1960II | L | Roosje | 573 | 600 | 128 | 1320 | 13 | 2880 | 2903 | 221 | 2090 | 11999 (37.3) | - 40 | - 184 | - 2244 | - 2362 | - 2303 |
| 45 | 41 | | 1960II | L | Jansje | 572 | 485 | 108 | 1109 | 13 | 2843 | 2845 | 219 | 2070 | 11446 (38.5) | - 13 | - 219 | - 2703 | - 2730 | - 2716 |
| 46 | 41 | | 1960II | E | Roosje | 541 | 791 | 166 | 1740 | 40 | 2764 | 2812 | 215 | 2029 | 12328 (45.8) | - 50 | - 91 | - 1085 | - 1435 | - 1260 |