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The Influence of Varying Maize Supplements on the Digestibility of the Cellulose in a Poor Veld Hay in Relation to the Bacterial Population of the Rumen of Sheep with a Note on the Nitrogen Metabolism.

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INTRODUCTION.

EXTENSIVE analytical evidence was produced by du Toit *et al* (1940) in studies on the natural veld that South African pastures are deficient not only in phosphate for the greater part of the year, but also in protein, more especially during winter or other times of food scarcity, such as drought. A glance at the analytical data published reveals extraordinary high values for the crude fibre content of the pastures analysed for the greater part of the year, suggesting the probability of a shortage of easily digestible, energy-producing material, particularly when stock are entirely dependent on mature, hard and fibrous grazing. This possible deficiency was investigated by du Toit, Malan and Smuts in a series of experiments with sheep, started in 1935 on the Government Farm, Nooitgedacht, in the Ermelo district situated in the Transvaal Highveld where frosts are severe during winter. A critical survey of the data produced in these experiments indicates that the supplementation of pasture with phosphate and protein during the winter months has little effect upon the condition of sheep unless the supplement contains carbohydrate as well, suggesting that poor use is being made of the energy-producing materials of the pasture under the existing conditions. Subsequently Smuts and Marais (1940) in a series of metabolism studies produced further evidence for the existence of an energy deficiency in the winter grazing of the Transvaal.

The energy-yielding part of the dry winter grazing consists almost exclusively of cellulose and other structural polysaccharides. An animal subsisting on such grazing would, therefore, be dependent upon its ability to utilise such carbohydrates for practically the whole of its energy requirements. It is consequently a matter of some interest, if not of practical importance to determine the magnitude of the cellulose utilization in winter grazing, and to ascertain to what extent, if at all, it is modified by supplementing the grazing with feeds necessary for the rectification of the known deficiencies, viz., protein, phosphate, and energy.

It is a well-known fact that the breakdown of cellulose and the hemicelluloses is accomplished not by enzymes secreted in the digestive tract but by enzymes of symbiotic micro-organisms. The quantitative relations involved in this microbiotic decomposition of carbohydrates are, however, suspected to be subject to variation depending upon the type and number of organisms present, which in turn are under the influence of the character of the food. Thus, it has been shown that the addition of easily digestible carbohydrates such as starch, cane sugar, or glucose to the ration of cattle or sheep reduced the digestibility of the fibre. The most recent evidence for this phenomenon has been supplied by Hamilton (1942) in experiments on sheep.

The degree of the breakdown of the structural carbohydrates is also intimately associated with their chemical and physical nature. It has often been demonstrated, for example by Louw (1942), that the complex polysaccharides of mature plants are not as well digested as they are in young, growing plants. The difference is due particularly to the presence of certain encrusting substances, notably lignin, which are deposited in increasing amount in the cell with advancing age. Micro-organisms have little or no action on lignin, especially on the lignin of mature plants, with the result that the cellulose is protected from the action of the organisms by the lignin or, probably, by a lignin-hemicellulose complex [c.f. Louw (Loc. cit.)]. Since it is known that pure isolated cellulose is almost completely digestible in the paunch of the ruminant it seems justifiable to state that in the presence of a sufficient number of the cellulose-digesting bacteria the digestion of the cellulose of mature plants (for instance, winter grazing in the Transvaal) will be limited largely by the degree of winter grazing in the Transvaal) will be limited largely by the degree of lignification. That the number and type of organisms may constitute yet another limiting factor, apart from lignification, in the digestion of the cellulose may be assumed from the work recently published by Harris and Mitchell (1941). These workers found that the addition of 5·0 grams urea to a basal ration containing only 0·136 per cent. nitrogen increased the digestibility of the cellulose from 17·8 to 38·7 per cent. They did not simultaneously study the influence of the urea supplement on the bacterial population of the rumen. However, data obtained at this Institute [van der Wath (1943)] show that the addition of urea to the type af basal ration employed by Harris and Mitchell does stimulate the proliferation of the paunch flora. Such an increase in the number of the organisms might conceivably have been responsible for the observed improvement in the digestion of the cellulose.

It has been found by one of us (J.G.v.d.W.) that the bacterial count in the rumen of sheep grazing on the natural veld of the Transvaal Highveld fluctuated with the changing condition of the pasture during the various seasons of the year. The highest count was found to occur during the summer months when the pasture is young, succulent, and of high nutritive value; the lowest count, on the other hand, was observed during the winter months when the veld is dry, fibrous, and of low nutritive value. Winter grazing in the Transvaal frequently has a protein content as low as 3·0 per cent. Conceivably, such a low protein content may limit the proliferation of the organisms in the rumen to such an extent that the breakdown of the cellulose cannot take place to the limit assumed to be set by the degree of lignification of the pasturage. The result would be that the animal is unable to utilise all the available energy-yielding nutrients of the winter grazing which is already deficient in protein and energy.

The experiments described below aimed primarily at ascertaining the influence of small supplements of protein-rich and carbohydrate-rich foodstuffs on the bacterial count in the rumen and on the digestibility of the cellulose in a ration of winter grazing containing approximately 3·0 per cent. of protein. Secondly, the carbohydrate-rich supplement was gradually increased to determine the effect on the microbiotic decomposition of the cellulose in the winter grazing. The nitrogen metabolism of the animals was studied in all the trials conducted.

Experimental.

The experiment consisted of a series of cellulose and nitrogen balance studies on five full-grown merino wethers with closed rumen fistulae and ranging from 75 to 130 lb. in body weight. Veld hay containing approximately 3·0 per cent. of protein, the type of feed which is representative of winter grazing, served as the basal ration and was fed in period 1. In period 2 each animal received daily in addition to the basal ration a quantity of meatmeal which was taken to be sufficient to rectify the deficit in the protein requirement for maintenance on the basis that an animal weighing 100 lb. requires about 24·0 grams of digestible protein per day for maintenance [c.f. Smuts and Marais (1939)]. From periods 3 to 7 the ration fed in period 2 was further supplemented with increasing amounts of crushed maize. The allowance of meatmeal was, however, reduced with each addition of maize in order to ensure a more or less constant intake of digestible protein from period 2 onwards. In period 7 when 300 grams of maize were fed and in the case of some of the lighter animals in earlier periods this level of protein intake was, however, more or less exceeded. During all the periods, including period 1 on the basal ration, each sheep received daily 3 grams of yeast, 5 grams of bone-ash, and 5 grams of common salt. Full details of the ration are given in Table 1. Unfortunately only two of the five sheep could be used in the first three periods. However, these periods were repeated with all five animals after the completion of period 7.

A preliminary feeding period of 10 days was allowed throughout except in the case of the repetition of period 3 following period 7 when the preliminary period lasted 15 days. Collection periods were of 10 days' duration. The Forbes type of metabolism cage was employed. The faeces and the urine were collected daily, the usual procedures for collecting, preserving, and aliquoting being followed. Feeds, faeces, and urine were analysed for total nitrogen by the usual Kjeldahl method. The method of Norman and Jenkins (1933) was employed to determine the cellulose in feeds and faeces. Samples of the ruminal ingesta were withdrawn through the fistulae on at least three consecutive days during a collection period for the bacterial counts and the average of such counts taken, the Petroff-Hanser counting chamber method described by van der Wath (1943) being used. The animals were fed twice daily, at 9 a.m. and 3 p.m., and the samples for the bacterial counts were withdrawn in the morning immediately before feeding.

Results.

(a) *Cellulose Digestion.*

The essential collection data are given in Table 1. Periods 1, 2, and 3 were repeated after the conclusion of period 7 in the order shown in the table. The discussion of the results will be based on the average values obtained in the seven periods in which five sheep were employed. The cellulose and protein contents of the feeds are presented in Table 2 while the coefficients of digestibility for cellulose together with the figures for the bacterial counts, representing millions of bacteria per cubic centimetre of ruminal ingesta, are given in Table 3.

In the light of a statistical analysis of the relevant data, which demands a difference of 169 between two means for significance at P = ·01, inspection of the averages for the five sheep given in the last column of Table 3 reveals two major changes in the bacterial count for consecutive periods of the experiment. The addition of the small amount of meatmeal to the basal ration resulted in a highly significant increase in the bacterial count from 1229 in period 1 (basal ration) to 1582 in period 2 (basal ration and meatmeal). The supplementation of the ration of period 2 with increasing amounts of crushed maize caused only minor increases in the bacterial count for consecutive periods up to and including period 5 for which a count of 1826 was obtained. A small decrease in the count to 1782 in period 6 was followed by a significant drop to 1566 in period 7, coinciding with an increase in the maize supplement from 200 to 300 grams per day (c.f. Table 1). It was observed that whereas for all other periods the bacterial counts made on consecutive days remained more or less constant a pronounced drop in the daily count occurred towards the end of period 7. Thus the counts for sheep 5 were 1760, 1445, 1225, and 1206 for the last four days of this period, respectively. Apparently, therefore, conditions in the rumen were most favourable for bacterial proliferation during periods 3, 4, and 5. The increase in the density of the bacterial population from periods 1 to 5 was associated with a progressive enrichment of the basal ration with highly digestible protein (meatmeal) and carbohydrate (maize). On the other hand further increases in the carbohydrate-rich supplement created conditions which became progressively less favourable to total bacterial growth. Towards the end of period 7 when the sudden decrease in the bacterial count was noted, the hydrogen ion concentration in the ruminal ingesta was determined and found to vary between 5·6 and 6·4 for the five animals. Although no pH determinations for any of the other periods were undertaken for comparison with these values there can be little doubt that the observed acidity was unfavourable to the growth of at least some of the bacteria.

Turning now to the average values for the digestibility of the cellulose given in the last column of Table 3 it is evident that the initial increases in the *number* of bacteria from periods 1 to 5 were not accompanied by an improvement in the digestibility of the cellulose. In fact, the tendency was rather in the opposite direction indicating an inverse relationship between the digestibility of the cellulose and the bacterial count for the first five periods of the experiment. In order to test the significance of the difference between the means for the coefficients of digestibility for cellulose in the seven periods of the experiment in which five sheep were employed the procedure for the well-known analysis of variance was applied. From this analysis it was established that the necessary difference between means for significance should be 3·30 for P = ·05, and 4·51, for P = ·01. Calculating from Table 3 it is found that the difference between the means for periods—

1 and 7 = 7·12, i.e., significant at P = ·01.
2 and 7 = 3·68, i.e., significant at P = ·05.
3 and 7 = 5·00, i.e., significant at P = ·01.
4 and 7 = 3·30, i.e., significant at P = ·05.
5 and 7 = 3·64, i.e., significant at P = ·05.
1 and 6 = 5·86, i.e., significant at P = ·01.
2 and 6 = 2·52, not significant.
3 and 6 = 3·74, i.e., significant at P = ·05.
1 and 5 = 3·48, i.e., significant at P = ·05.
1 and 4 = 3·82, i.e., significant at P = ·05.

Differences not specified in the above summary were not significant. The general tendency for the digestibility of the cellulose was to decrease gradually from period 1 to 7. That being so, it may be expected that if the difference between periods 3 and 6 is significant, that between periods 2 and 6 should show even greater significance. In reality the difference between the latter two periods was found to be insignificant. At this juncture no feasible explanation can be offered for this finding except that it may be in the sequence of the seven periods on which the statistical analysis was made—periods 3, 2, and 1 following in this order after period 7. Inspection of the individual coefficients of digestibility reveals that all five animals digested the cellulose less efficiently in period 2 (basal ration + meatmeal) than in either period 3 (basal ration + meatmeal + maize) or period 1 (basal ration).

However that may be, the salient feature in the results remains that the digestibility of the cellulose in the hay, representing poor winter grazing, was not improved by supplementing the hay with varying amounts of meatmeal and crushed maize in spite of an increase in the number of bacteria in the rumen—the site of cellulose digestion.

At this stage it should, however, be pointed out that the method employed yields figures for the total number of bacteria in the ruminal ingesta. The organisms counted were not all of the same kind and it is conceivable that some of the species were not essential in the process of cellulose digestion. The number of the organisms actually responsible for the breakdown of the cellulose molecule may have undergone no change at all, it may have increased, or it may even have decreased with the modifications in the rations fed in periods 1 to 7. Whatever the case may be, it seems warranted to infer that the ingestion of the basal ration, representing, approximately, the nutritive value of winter grazing in the Transvaal, created conditions in the rumen of the sheep sufficiently favourable to the existence of that number of organisms of the right kind necessary for the maximum utilization of the cellulose in the ration. If that be so, then the degree of lignification, which, as pointed out earlier, is known to influence cellulose digestion, seems to be an important if not the only factor governing the magnitude of the breakdown of the cellulose in pasturage containing about 3·0 per cent. protein. The qualification as to protein content (N x 6·25) is considered pertinent in view of the recent finding of Harris and Mitchell (loc. cit.), previously referred to, viz., that the digestion of the cellulose in a N-low ration can be improved by supplementing such a ration with a nitrogenous substance. As stated, the basal ration employed by these workers had a nitrogen content of only 0·136 per cent. or 0·85 per cent. protein. Van der Wath (loc. cit.), found that the bacterial count in the rumen of sheep on a basal ration containing 0·3 per cent. nitrogen increased from 612 to 1068 when the basal ration was supplemented daily with 2·33 grams nitrogen in the form of urea, and from 612 to 1875 when a similar daily supplement of nitrogen was given in the form of white fishmeal. The influence of these increases on cellulose digestion was, unfortunately, not studied. Nevertheless, van der Wath's results in conjunction with the results obtained by Harris and Mitchell seem to justify the conclusion that the total number of ruminal bacteria as determined in this investigation will constitute a second limiting factor in the digestion of the cellulose in rations very low in nitrogen content, in addition to the well-known influence of the degree of lignification of the plant material.

The depressing influence of the greater maize supplements on the digestibility of the cellulose in the basal ration has previously been indicated. The magnitude of this depression in digestibility was governed by the size of the supplement so that the odds in favour of the significance of the depression increased generally as the supplements of maize increased. The digestibility of the total dry matter improved on an average from 47·5 per cent. in period 1 to 60·7 per cent. in period 7, corresponding with an increasing amount of highly digestible maize in the total dry matter eaten (c.f. Table 1). Inspection of the mean values given in the last column of Table 1 reveals, however, that the dry matter of the veld hay alone, which contributed practically the whole of the cellulose in the supplemented rations, was digested in a manner similar to the cellulose in periods 1 to 7, decreasing from 47·5 per cent. in period 1 to 36·1 per cent. in period 7, These coefficients which were calculated on the assumption that the maize was completely digestible somewhat exaggerate the depressing influence of the added maize on the digestibility of the hay. Odd pieces of undigested maize have, for instance, occasionally been detected in the faeces, especially in that collected in periods 6 and 7 when the heavier supplements were given. Nevertheless, such a depression in digestibility is in agreement with a fact established by numerous experiments (c.f. Armsby, 1917). The effect is most distinct when pure digestible carbohydrates, such as starch, cane sugar, etc., are added, but manifests itself also when large amounts of feeding stuffs rich in carbohydrates are introduced. In the latter case it is, however, often impossible to follow the quantitative relations clearly. Thus, in the experiment under discussion it is not possible to determine what proportion of the total faecal dry matter was derived from the maize and what from the veld hay. However, the undigested cellulose present in the faeces may be taken to be derived almost exclusively from the veld hay, due to the very small contribution of the maize to the total intake of this constituent. For this reason the magnitude of the depression on the digestibility of the dry matter of the veld hay, caused by the maize supplements, seems to be best reflected in the coefficients of digestibility obtained for cellulose.

To conclude this discussion reference may be made to the possible influence of the supplement necessary for the rectification of the known deficiencies for maintenance in winter grazing, mentioned in the introduction, on the utilization of its available energy. Smuts and Marais (loc. cit.) inferred from a series of metabolism studies with sheep that 150 grams of maize daily will successfully supplement the protein and energy deficiencies for maintenance in winter grazing. The basal ration of poor veld hay was supplemented with this amount of maize in period 5 of the present investigation. Reference to Table 3 reveals that the 150 grams of maize depressed the digestibility of the cellulose in the basal ration from 65·5 to 62·1 per cent. Although the odds in favour of this difference being significant was found to be 100:5 the actual depression in digestibility may be considered to be of no practical importance in the energy metabolism of the animal.

(*b*) *Nitrogen Metabolism.*

The data relating to the nitrogen metabolism of the five sheep during the seven periods of the experiment are presented in Table 4. The mean values on which the following brief discussion is based are given in the last column of the table.

In period 1 on the basal ration the daily nitrogen intake amounted to 2·48 grams of which only 0·23 gram was apparently digested. The animals were definitely not receiving sufficient nitrogen for maintenance as was evidenced by the pronounced negative balance of 1·03 grams nitrogen per day. A protein deficiency of this nature has been shown to prevail for almost six months of the year in certain areas of the Union of South Africa [c.f. du Toit *et al* (1940) and Smuts and Marais (1940)]. In period 2 the total daily nitrogen intake increased to 5·87 grams, mainly due to the supplement of meatmeal. The nitrogen apparently digested increased ten-fold to 2·31 grams, but in spite of this the nitrogen balance remained negative at 0·93 grams per day. From period 2 to period 6 the daily nitrogen intake remained practically the same, the small increases being mainly due to the slightly higher nitrogen content of the hay consumed. The nitrogen apparently digested increased relatively more, from 2·31 grams in period 2 to 2·85 grams in period 6. The small increases in digestible nitrogen intake was accompanied by marked changes in the manner of its utilization. Thus, while the amount of nitrogen excreted in the faeces remained more or less the same, fluctuating between 3·33 and 3·63 grams daily, that excreted in the urine decreased from 3·24 grams daily in period 2 to 1·90 grams daily in period 6. Simultaneously the daily nitrogen balance changed from −0·93 in period 2 to +0·95 in period 6, i.e., from a relatively strong negative to a relatively strong positive nitrogen balance. Reference to Table 4 shows that meatmeal nitrogen was gradually replaced by nitrogen derived from maize in the rations fed from periods 2 to 6. From this it may be inferred, either that the protein of meatmeal was not as efficiently utilized as that of maize, or that portion of the protein was catabolized for energy purposes especially in period 2. The effect of both these metabolic processes would be that less protein was available for fulfilling the nitrogen requirements for maintenance. The first-mentioned possibility is, however, ruled out by the biological values obtained for the proteins of the two foodstuffs, viz., 67·0 for maize [Marais and Smuts (1940)] and the same figure, 67·0, for meatmeal [du Toit and Smuts (1941)]. If, at the same time, it is remembered that the gradual replacement of meatmeal nitrogen by maize nitrogen was unavoidably accompanied by the introduction of an increasing amount of highly digestible carbohydrate to the rations fed in periods 2 to 6 (c.f. Table 1), then the only feasible explanation for the observed changes in the nitrogen utilization seems to be that an energy deficiency existed in at least the ration of period 2. The increasing supplements of maize from period 3 onwards gradually eliminated the energy deficiency with the result that the available nitrogen could more and more be utilized for its primary function in metabolism, viz., the replenishment of the unavoidable nitrogen losses associated with the minimum metabolism of the protoplasm, and tissue growth.

Similar results have been obtained by Smuts and Marais (loc. cit.). They found, for instance, that when winter grazing was supplemented with 56·0 grams of peanutmeal per day the sheep were in a considerably negative nitrogen balance, in spite of the fact that the nitrogen intake was raised to 6·5 grams daily by the supplement. On the other hand, in another trial where straw replaced the winter grazing, the daily allowance of peanutmeal reduced to 32·0 grams, and sufficient energy provided in the form of dextrinized starch, the sheep were found to be on the whole in nitrogen equilibrium with a daily nitrogen intake of only 3·5 grams. The conclusion drawn by these workers, viz., that " under practical conditions it will be futile to rectify the existing protein deficiency with a minimum quantity of protein unless the energy requirements are (simultaneously) satisfied " is strongly supported by the results of the present investigation.

Summary.

From the results of a series of metabolism studies on sheep with open rumen fistulae in which a basal ration of winter grazing was supplemented with meatmeal and increasing amounts of crushed maize it was found that:—

(1) Small amounts of meatmeal and supplements of maize ranging from 50 grams to approximately 150 grams per day favoured the growth of the rumen organisms. Heavier supplements of maize, on the other hand, tended to reduce the number of organisms in the rumen.

(2) The increase in the bacterial count did not improve the digestibility of the cellulose in the winter grazing. A progressive depression in its digestibility with increasing supplements of maize was, however, observed.

(3) The rectification of the existing protein deficiency in winter grazing with a minimum quantity of protein is futile unless its energy deficiency is simultaneously satisfied.

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TABLE 1.

Showing Collection Data and Digestibility of Dry Matter.

Period.	Sheep No.	Weight (lb.).		Daily Ration (Gm.).			Average Daily Orts.* (gm. Dry Matter).	Average Daily Dry Matter Intake (gm.).		Average Daily Dry Matter Excreted. (gm.).	Coefficient of Digestibility for dry matter.			
		Initial.	Final.	Hay.	Maize.	Meat-meal.		Hay.	Total.		Total.	Average.	Hay.	Average.
1	3	100	99	600	0	0	33·0	538·0	538·0	293·4	45·5	—	45·5	—
	5	135	133	600	0	0	11·0	561·0	561·0	279·4	50·2	—	50·2	—
2	3	99	97	600	0	25	21·4	532·4	556·2	289·1	48·0	—	45·7	—
	5	129	127	600	0	34	14·5	539·3	571·6	286·8	49·8	—	46·9	—
3	3	93	94	600	50	17	21·2	544·8	606·5	299·1	50·7	—	45·1	—
	5	125	125	600	50	28	8·8	557·2	629·4	280·0	55·6	—	49·8	—
4	3	95	95	600	100	10	3·9	551·1	651·8	306·3	53·0		44·4	
	4	108	109	600	100	16	45·0	510·0	616·4	281·3	54·4		44·8	
	5	125	128	600	100	20	1·0	554·1	664·3	290·8	56·2	51·9	47·6	42·6
	6	92	86	600	100	10	18·5	536·5	637·2	314·0	50·8		41·5	
	7	75	74	600	100	4	59·5	495·5	590·5	324·8	44·9		34·5	
5	3	95	96	600	150	4	13·0	539·0	679·6	306·2	55·0		43·2	
	4	112	110	600	150	7	66·8	485·2	628·6	249·6	60·3		48·6	
	5	132	131	600	150	12·5	0	552·0	700·7	270·0	61·5	56·0	51·1	43·9
	6	85	87	600	150	4	41·6	510·4	651·0	300·9	53·8		41·1	
	7	76	78	600	150	0	55·0	497·0	633·8	321·3	49·4		35·4	
6	3	96	96	600	200	0	76·0	476·0	658·4	279·7	57·5		41·2	
	4	110	111	600	200	1·5	79·9	472·1	655·9	265·3	59·6		43·8	
	5	131	132	600	200	7·0	3·7	548·3	737·3	304·6	58·7	56·8	44·4	41·0
	6	88	87	600	200	0	33·1	518·9	701·3	313·9	55·3		39·5	
	7	79	77	600	200	0	39·4	512·6	695·0	328·8	52·7		35·9	

* Orts were composed of hay only.

TABLE 1.—(*continued*).

Period.	Sheep No.	Weight (lb.).		Daily Ration (Gm.).			Average Daily Orts.* (gm. Dry Matter).	Average Daily Dry Matter Intake (gm.).		Average Daily Dry Matter Excreted. (gm.).	Coefficient of Digestibility for dry matter.			
		Initial.	Final.	Hay.	Maize.	Meat-meal.		Hay.	Total.		Total.	Average.	Hay.	Average.
7	3	97	96	600	300	0	166·0	383·0	656·6	239·8	63·5		37·4	
	4	112	113	600	300	0	192·0	357·0	630·6	230·8	63·4		35·4	
	5	135	136	600	300	0	33·6	515·4	789·0	289·0	63·4	60·7	44·0	36·1
	6	91	88	600	300	0	76·0	473·0	746·6	316·5	57·6		33·1	
	7	79	78	600	300	0	60·1	488·9	762·5	340·6	55·4		30·4	
3† (Rep.).	3	99	97	600	50	16	20·2	524·6	585·4	282·9	51·6		46·0	
	4	110	110	600	50	19	109·0	435·8	499·4	221·2	55·7		49·3	
	5	128	127	600	50	27	5·4	539·4	610·6	285·9	53·2	50·7	47·0	44·7
	6	86	86	600	50	15	25·6	519·2	579·0	309·2	46·6		40·5	
	7	78	76	600	50	12	23·5	521·3	578·3	309·2	46·6		40·7	
2 (Rep.)	3	99	95	600	0	24	26·5	521·3	544·1	288·0	47·0		44·8	
	4	109	106	600	0	25	119·1	428·7	452·4	230·6	49·1		46·2	
	5	128	121	600	0	33	75·6	472·2	503·6	269·4	46·5	45·0	43·0	42·4
	6	87	83	600	0	21	30·8	517·0	537·0	323·8	39·7		37·4	
	7	77	76	600	0	18	28·1	519·7	536·8	308·2	42·6		40·7	
1 (Rep.)	3	89	90	600	0	0	107·5	438·5	438·5	232·3	47·0		47·0	
	4	102	104	600	0	0	122·6	423·4	423·4	203·5	51·9		51·9	
	5	117	117	600	0	0	105·5	440·5	440·5	233·1	47·1	47·5	47·1	47·5
	6	81	83	600	0	0	54·3	491·7	491·7	265·1	46·1		46·1	
	7	83	75	600	0	0	37·5	508·5	508·5	278·2	45·3		45·3	

† Repetition.

TABLE 2.

Showing Percentage Composition of Feeds (Dry Matter Basis).

Feed.	Constituent.	Periods.									
		1	2	3	4	5	6	7	3 (Rep.).	2 (Rep.).	1 (Rep.).
Hay.....	Cellulose.....	51·3	52·3	50·4	49·9	48·8	48·6	48·4	48·7	48·7	49·5
	Crude protein	3·00	3·24	3·11	3·55	3·49	3·41	3·42	3·16	3·16	3·00
Maize....	Cellulose.....						1·8				
	Crude protein						11·0				
Meatmeal.	Crude protein						80·0				
Yeast....	Crude protein						50·0				

TABLE 3.

Showing Coefficients of Digestibility for Cellulose with the Ruminal Bacterial Counts in Brackets.*

Period.	Sheep 3.	Sheep 4.	Sheep 5.	Sheep 6.	Sheep 7.	Means.
1............	62·9 (1148)	—	68·1 (926)	—	—	—
2............	66·2 (1844)	—	67·4 (1696)	—	—	—
3............	63·7 (1864)	—	68·8 (1900)	—	—	—
1 (Repetition)..	65·4 (1200)	71·4 (1189)	65·2 (1156)	63·6 (1244)	62·1 (1356)	65·5 (1229)
2 (Repetition)..	63·2 (1511)	66·7 (1644)	64·7 (1711)	56·6 (1500)	59·8 (1542)	62·2 (1582)
3 (Repetition)..	65·3 (1644)	69·5 (1756)	67·7 (1722)	57·1 (1622)	57·5 (1767)	63·4 (1702)
4............	63·9 (1889)	65·3 (1789)	67·7 (1566)	59·6 (1839)	52·1 (1844)	61·7 (1785)
5............	61·9 (1785)	67·6 (1907)	70·5 (1700)	58·2 (1781)	52·1 (1948)	62·1 (1826)
6............	59·3 (1800)	64·5 (1944)	66·4 (1744)	57·1 (1733)	51·1 (1689)	59·7 (1782)
7............	58·9 (1517)	61·2 (1622)	67·7 (1408)	54·0 (1644)	50·3 (1641)	58·4 (1566)

* Figures represent millions of bacteria per c.c. ruminal contents.

TABLE 4.

Nitrogen Metabolism of Sheep in Periods 1 to 7 (1 to 3 were Repetitions).

Period.		Sheep 3.	Sheep 4.	Sheep 5.	Sheep 6.	Sheep 7.	Average for 5 Sheep.
1 (Rep.)	Nitrogen intake :—						
	Hay.........................	2·01	2·23	2·02	2·45	2·51	2·24
	Yeast.......................	·24	·24	·24	·24	·24	·24
	TOTAL............	2·25	2·47	2·26	2·69	2·75	2·48
	Nitrogen outgo :—						
	Faeces........................	2·11	2·07	2·18	2·41	2·50	2·25
	Urine.........................	1·01	1·41	1·41	1·27	1·18	1·26
	TOTAL............	3·12	3·48	3·59	3·68	3·68	—
	N.—Balance....................	−0·87	−1·01	−1·33	−0·99	−0·93	*−1·03*
	N.—(Apparently) digested.......	0·14	0·40	0·08	0·28	0·25	0·23
	%N.—(Apparently) digested.....	6·2	16·2	3·5	10·4	9·1	9·1

TABLE 4.—(*continued*).

Period		Sheep 3.	Sheep 4.	Sheep 5.	Sheep 6.	Sheep 7.	Average for 5 Sheep.
2 (Rep.)	Nitrogen intake :—						
	Hay	2·64	2·38	2·39	2·61	2·63	2·53
	Yeast	·24	·24	·24	·24	·24	·24
	Meatmeal	3·07	3·20	4·22	2·69	2·30	3·10
	TOTAL	5·95	5·82	6·85	5·54	5·17	5·87
	Nitrogen outgo :—						
	Faeces	3·56	3·17	3·77	3·78	3·50	3·56
	Urine	2·88	3·60	3·99	2·94	2·80	3·24
	TOTAL	6·44	6·77	7·76	6·72	6·30	—
	N.—Balance	−0·49	−0·95	−0·91	−1·18	−1·13	*−0·93*
	N.—(Apparently) digested	2·39	2·65	3·08	1·76	1·67	2·31
	%N.—(Apparently) digested	40·2	45·6	45·0	31·8	32·3	39·0
3 (Rep.)	Nitrogen intake :—						
	Hay	2·65	2·39	2·73	2·63	2·64	2·61
	Yeast	·24	·24	·24	·24	·24	·24
	Meatmeal	2·05	2·43	3·45	1·92	1·53	2·27
	Mealies	·81	·81	·81	·81	·81	·81
	TOTAL	5·75	5·87	7·23	5·60	5·22	5·93
	Nitrogen outgo :—						
	Faeces	3·30	2·88	3·75	3·37	3·33	3·33
	Urine	1·88	3·04	3·00	2·22	2·18	2·46
	TOTAL	5·18	5·92	6·75	5·59	5·51	—
	N.—Balance	+0·57	−0·05	+0·48	+0·01	−0·29	*+0·14*
	N.—(Apparently) digested	2·45	2·99	3·48	2·23	1·89	2·61
	%N.—(Apparently) digested	42·6	50·9	48·2	39·8	36·2	43·5
4	Nitrogen intake :—						
	Hay	3·13	3·00	3·15	3·05	2·95	3·06
	Yeast	·24	·24	·24	·24	·24	·24
	Meatmeal	1·28	2·05	2·56	1·28	·51	1·53
	Mealies	1·61	1·61	1·61	1·61	1·61	1·61
	TOTAL	6·26	6·90	7·56	6·18	5·31	6·44
	Nitrogen outgo :—						
	Faeces	3·62	3·64	3·77	3·57	3·55	3·63
	Urine	2·16	3·37	3·09	2·71	2·17	2·70
	TOTAL	5·78	7·01	6·86	6·28	5·72	—
	N.—Balance	+0·48	−0·11	+0·70	−0·10	−0·41	*+0·11*
	N.—(Apparently) digested	2·64	3·26	3·79	2·61	1·76	2·81
	%N.—(Apparently) digested	42·2	47·3	50·2	42·3	33·1	43·0

TABLE 4.—(*continued*).

Period.		Sheep 3.	Sheep 4.	Sheep 5.	Sheep 6.	Sheep 7.	Average for 5 Sheep.
5	Nitrogen intake :—						
	Hay	3·00	2·86	3·08	2·94	2·90	2·96
	Yeast	·24	·24	·24	·24	·24	·24
	Meatmeal	·51	·90	1·60	·51	0	·70
	Mealies	2·42	2·42	2·42	2·42	2·42	2·42
	TOTAL	6·17	6·42	7·34	6·11	5·56	6·32
	Nitrogen outgo :—						
	Faeces	3·60	3·24	3·53	3·47	3·52	3·47
	Urine	1·64	2·17	2·63	2·17	1·68	2·06
	TOTAL	5·24	5·41	6·16	5·64	5·20	—
	N.—Balance	+0·93	+1·01	+1·18	+0·47	+0·36	*+0·79*
	N.—(Apparently) digested	2·57	3·18	3·81	2·64	2·04	2·85
	%N.—(Apparently) digested	41·7	49·6	51·9	43·2	36·7	44·6
6	Nitrogen intake :—						
	Hay	2·46	2·76	3·00	2·84	2·80	2·77
	Yeast	·24	·24	·24	·24	·24	·24
	Meatmeal	0	·19	·90	0	0	·22
	Mealies	3·22	3·22	3·22	3·22	3·22	3·22
	TOTAL	5·92	6·41	7·36	6·30	6·26	6·45
	Nitrogen outgo :—						
	Faeces	3·29	3·51	3·92	3·60	3·68	3·60
	Urine	1·48	1·99	2·77	1·71	1·54	1·90
	TOTAL	4·77	5·50	6·69	5·31	5·22	—
	N.—Balance	+1·15	+0·91	+0·67	+0·99	+1·04	*+0·95*
	N.—(Apparently) digested	2·63	2·90	3·44	2·70	2·58	2·85
	%N.—(Apparently) digested	44·5	45·2	46·8	42·9	41·2	44·1
7	Nitrogen intake :—						
	Hay	2·04	2·37	2·81	2·75	2·80	2·55
	Yeast	·24	·24	·24	·24	·24	·24
	Mealies	4·84	4·84	4·84	4·84	4·84	4·84
	TOTAL	7·12	7·45	7·89	7·83	7·88	7·63
	Nitrogen outgo :—						
	Faeces	3·58	3·67	3·97	4·23	4·40	3·97
	Urine	1·55	1·98	2·11	1·74	1·76	1·83
	TOTAL	5·13	5·65	6·08	5·97	6·16	—
	N.—Balance	+1·99	+1·80	+1·81	+1·86	+1·72	*+1·84*
	N.—(Apparently) digested	3·54	3·78	3·92	3·60	3·48	3·66
	%N.—(Apparently) digested	49·7	50·7	49·7	46·0	44·2	48·1