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Comparison of grass silage utilization by reindeer and sheep.

2. Rumen fermentation and rumen microbiota

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Abstract. The criteria used in comparing the utilization of grass silage by reindeer and sheep were rumen pH, ammonia, volatile fatty acids (VFA) and microbes.

Rumen samples were taken before feeding, and 2½ and 5½ hours after the beginning of feeding. Rumen fermentation was lower in the reindeer than in the sheep and differed less between the three sampling times. In the reindeer/the pH of the rumen fluid averaged 6.94 and in the sheep 6.61. The average amounts of NH_3–N were 17.0 and 24.2 mg/100 ml rumen fluid and those of total VFA 8.46 and 10.90 mmoles/100 ml rumen fluid, respectively. The proportion of acetic acid in the VFA in the reindeer was 75.3 molar % and in the sheep 66.0 molar %, the corresponding values for propionic acid being 18.5 and 22.0 molar % and for butytic acid 4.2 and 8.8 molar %.

The number of rumen ciliates in the reindeer averaged 87/mm^3 rumen contents and in the sheep 314/mm^3. The numbers of bacteria were 16.0 × 10^6/mm^3, respectively. The proportion of the total microbe mass in the reindeer rumen contents was 1.8 % and in the sheep 2.4 %. The proportions of bacteria in this mass were 87 % and 70 %, respectively. The differences between the reindeer and sheep in the rumen fermentation results and in the numbers of rumen microbiota were nearly all statistically significant ($P<0.001$ – $P<0.05$).

Introduction

The principal products of rumen fermentation are ammonia and volatile fatty acids (VFA). Both amounts of these products and the rumen microbial synthesis have been used as criteria, when describing feed utilization by ruminants. Considerable research has thus been devoted to the dietary factors which affect their amounts and to the proportions of the individual fatty acids. The domestic ruminant used most often in these experiments has been the sheep. Being only a semi-domestic animal the reindeer has less frequently been chosen for this kind of physiological study.

The main purpose of the present study was to investigate grass silage as a feed for reindeer and to compare its utilization between reindeer and sheep.

This paper deals with the rumen fermentation and rumen microbes in reindeer and sheep on silage diets. The palatability and nutritive value of silage have been compared between the two animals in another paper (SYRJÄLÄ-QVIST 1982).

Experimental procedures

The experiment was performed with three adult male reindeer and three adult Finsheep rams, using a 3 × 3 Latin square design. Grass silage was the only feed. The experimental procedures are explained in more detail in the earlier paper (SYRJÄLÄ-QVIST 1982).

Rumen samples were taken three times in each of the last two days of every experimental period. They were obtained from the sheep through a permanent rumen fistula and from the reindeer through the mouth with soft silicone tube, 1.5 cm in diameter. The sampling times were: before feeding in the morning, and 2 1/2 and 5 1/2 hours after the beginning of feeding. For pH, ammonia and VFA the sampling and the treatments and analyses of the samples were the same as described by SYRJÄLÄ (1972), and for the rumen microbiota they were as described by SYRJÄLÄ et al. (1973, 1976). The pH was measured, and the ammonia and rumen microbes determined on each sample, altogether 48 samples from the reindeer and 54 from the sheep. For the determination of volatile fatty acids, the samples taken at the same sampling time on the two days were mixed together.

The silages were prepared from grass at three different stages of growth. As the chemical composition and the quality of the different silages were the same (Table 1, SYRJÄLÄ-QVIST 1982), the rumen fermentation results are expressed as the average values for all the silages.

Results and discussion

Rumen fermentation

pH. At every sampling time the pH values of the rumen contents of the reindeer were significantly higher than those of the sheep ($P < 0.05$ or $P < 0.001$), the average values for the reindeer at the different times being 7.08, 6.86 and 6.88, as opposed to 6.77, 6.50 and 6.56 for the sheep (Fig. 1, Table 2). The individual differences were larger in the reindeer than in the sheep. A reason for the higher pH values for the reindeer may be the different sampling technique. Being taken through the mouth the rumen samples from the reindeer, may possibly have been contamined by saliva, although the tube was rinsed with rumen contents before collection.

In an experiment where the reindeer were rumen-fistulated, the pH of the rumen contents on silage feeding was 6.35–6.70 and on lichen feeding 6.15–6.20 (SYRJÄLÄ 1978). The pH values for the sheep in the present

Table 1. The chemical composition and quality of the silage.

	Mean	Variation between the growth stages
Dry matter, %	25.9	25.2–26.8
% of dry matter:		
Ash	11.2	10.4–12.6
Crude protein	15.0	14.2–16.3
True protein	9.8	8.7–11.1
Crude fat	5.6	5.2–6.1
N-free extrack	40.4	39.5–41.5
Crude fibre	27.8	27.1–28.5
Sugars as glucose	7.9	5.8–10.4
% of total N:		
Ammonia N	4.1	3.8–4.4
Soluble N	51	51
pH	3.82	3.80–3.85

Table 2. pH, ammonia nitrogen and volatile fatty acids in the rumen fluid of reindeer and sheep. The values are the averages of the different sampling times.

	Reindeer	Sheep
pH	6.94	6.61
NH_3–N, mg/100 ml	17.0	24.2
Total VFA, mmoles/100 ml	8.46	10.90
Acetic acid, molar %	75.3	66.0
Propionic acid "	18.5	22.0
Butyric acid "	4.2	8.8
Isovaleric acid "	1.6	2.3
Valeric acid "	0.4	0.9
Ratio acetic: propionic	4.1	3.0
" acetic: butyric	18.9	7.7
" propionic: butyric	4.6	2.5

experiment were at the same level as in another experiment with animals on silage feeding (SYRJÄLÄ 1972).

Ammonia. The ammonia in the rumen fluid of the reindeer and sheep was at the same level in the samples taken before feeding (Fig. 1, Table 2), but after feeding it rose more sharply in the sheep, the increase being significant ($P < 0.01$). The average ammonia values for the reindeer at the different sampling times, 16.8, 18.9 and 15.2 NH_3-N mg/100 ml of rumen fluid, were quite near the values in the earlier silage experiment, where they were 14.7–16.4 mg/100 ml (SYRJÄLÄ 1978), but higher than those recorded on lichen feeding, which were 5.6–6.8 mg/100 ml. The ammonia values for the sheep correspond to those in the earlier study (SYRJÄLÄ 1972).

The fact that the values for the reindeer differed so little between the sampling times can mainly be explained by their eating habits. The reindeer ate their silage rations very slowly during the whole day, whereas the sheep finished them in about half an hour. Similar eating habits have been observed

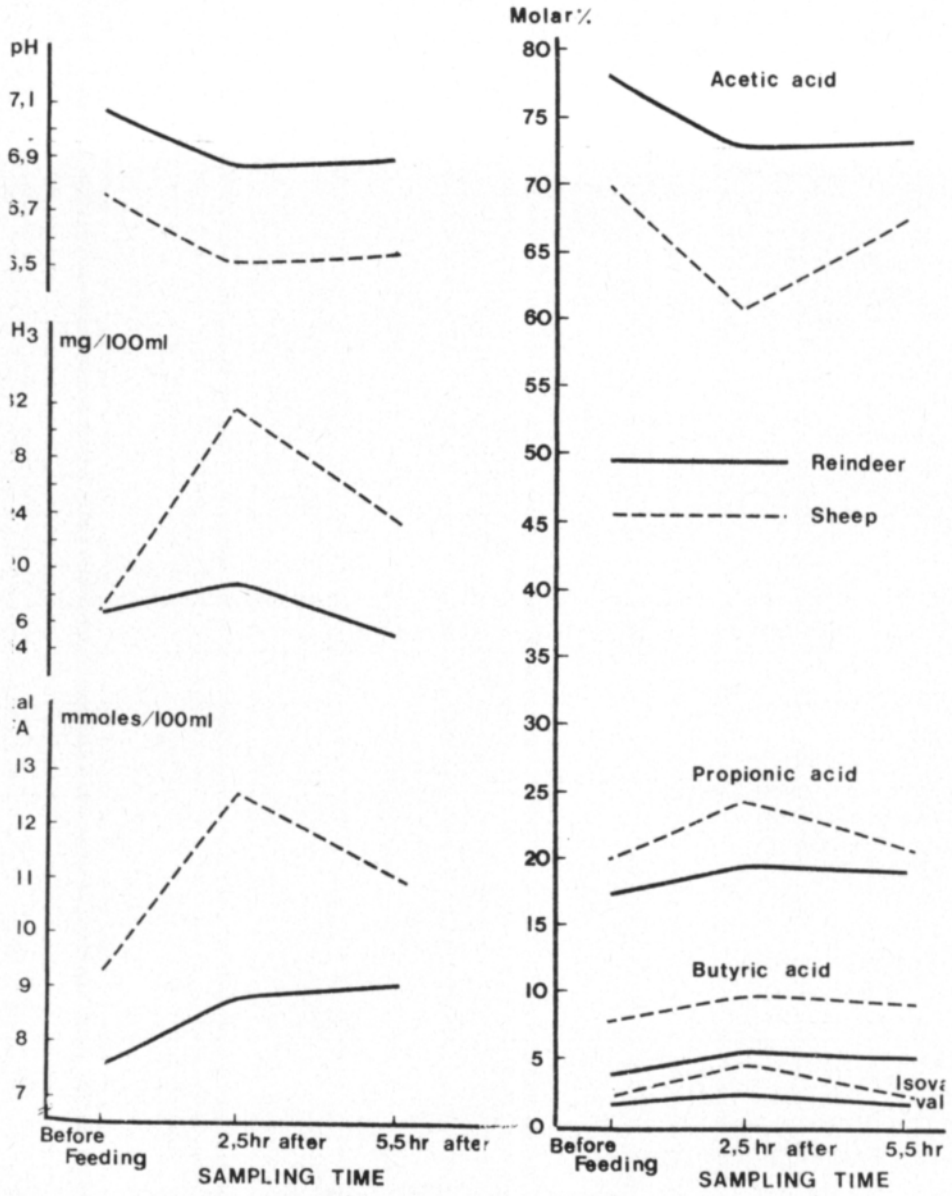

Fig. 1. pH, NH_3–N and VFA in the rumen fluid of reindeer and sheep.

in an earlier experiment with reindeer on silage feeding (SYRJÄLÄ and HEIKKILÄ 1975) and also when silage was fed in practical reindeer husbandry (KURKELA 1976).

The amount of silage consumed each day by the reindeer in this experiment was about half of that eaten by the sheep, 3.6 kg versus 7.1 kg (SYRJÄLÄ-QVIST 1982). The total water intake by the reindeer was 5 kg/kg DM eaten, and by the sheep 3 kg.

Volatile fatty acids. The total amount of VFA was not determined separately, but was taken as being the sum of the amounts of acetic,

propionic, butyric, isovaleric and valeric acids, which were determined by gas chromatographic analyses and expressed as mmoles/100 ml of rumen fluid (see SYRJÄLÄ 1972).

The total VFA in the rumen fluid of the reindeer was significantly lower ($P<0.001$ – $P<0.05$) at every sampling time than in that of the sheep (Fig. 1, Table 2). The values for the reindeer at the different sampling times were 7.54, 8.85 and 9.00 mmol/100 ml, and did not differ significantly from each other ($P>0.05$), whereas the values for the sheep differed significantly ($P<0.05$ or $P<0.01$), being 8.21, 12.51 and 10.98 mmol/100 ml.

The variation between the sampling times in the proportions of the different fatty acids was similar in the reindeer and the sheep (Fig. 1). The proportion of acetic acid in the total VFA of the reindeer was 73–78 mol % and the values for the sheep were 61–70 mol %; the proportions of propionic acid were 18–19 and 20–25 mol % and those of butyric acid 3–5 and 8–9 mol %, respectively.

In an earlier experiment with reindeer on a silage diet (SYRJÄLÄ 1978), the total VFA of the rumen fluid was higher than in this experiment, averaging 12.3 mmol/100 ml. On a lichen diet it averaged 9.8 mmol/100 ml. The proportion of acetic acid in the total VFA was about 79 mol % on the silage diet and 68 mol % on the lichen diet.

The total VFA in the sheep were a little higher than in another study with sheep on silage feeding (SYRJÄLÄ 1972). There were also some differences in the proportions of the individual fatty acids, that of acetic acid being lower and those of the other fatty acids being higher than in the earlier study.

Rumen microbiota

Number and kinds of ciliate and bacteria cells

In both the reindeer and sheep the numbers of ciliate and bacteria cells depended on the sampling being highest before feeding (Fig. 2–3). In this study, however, attention will mainly be directed to the average values of the different sampling times, so that the difference in the eating habits of the reindeer and sheep will not interfere so much with the comparison between the two animals. When the results of this experiment are compared with those in the literature, the values of the samples taken before feeding will be used, because in most of the earlier studies the samples were taken before feeding or as long as some days after feeding.

Ciliates. The total number of ciliate cells in the reindeer was less than one third of that in the sheep ($P<0.001$, Table 3, Fig. 2). The value for the sheep before feeding was 407 ciliates/mm^3, being at nearly the same level as the value recorded in another experiment where the sheep were on silage feeding (SYRJÄLÄ et al. 1976). The corresponding value for the reindeer in the present experiment was 109 ciliates/mm^3. On a natural diet in winter, it varied from 188 to 1183/mm^3, depending on the food supply (SYRJÄLÄ et al. 1973). In the experiment of WESTERLING (1970) it was as high as 556–1450/mm^2 even in poor nutritional conditions.

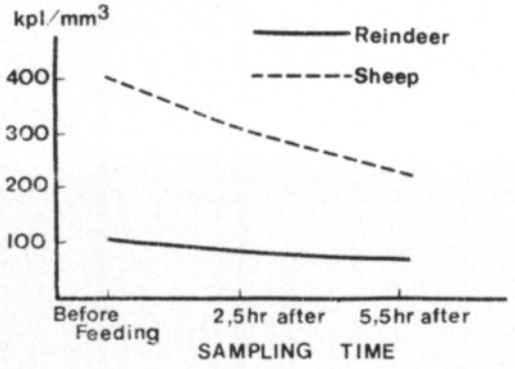

Fig. 2. The numbers of ciliate cells at the different sampling times.

Table 3. The number of ciliate and bacteria cells per mm^3 rumen contents and the percentage composition of the ciliate fauna. The values are the averages of the different sampling times.

	Numbers		Percentage composition of fauna	
	Reindeer	Sheep	Reindeer	Sheep
Ciliates, total	87	314	100	100
Holotrica	3	44	3.1	14.2
Entodinium	74	251	85.1	80.1
Diplodinium	5	7	6.0	2.3
Eudiplodinium	4	8	5.3	2.5
Ostracodinium	0–1	2	0.5	0.7
Enoploplastron	0	1	0.0	0.2
Bacteria × 10^6	16.0	17.4		

Of the Holotrichs, only *Dasytricha ruminantium* was found in the reindeer, as also in earlier experiments (WESTERLING 1970, SYRJÄLÄ et al. 1973). According to the review of GIESECKE (1970) concerning the rumen protozoa of different animals, *Dasytricha* is not found at all in the rumen of reindeer. *Isotrica prostoma* was also found in the sheep, although it was lacking in many samples. In contrast, *Charon equi* was fairly common in the sheep.

Of the Entodinimorphs, the species of the genera *Entodinium* were the most common, accounting for more than 80 % of the ciliate numbers in both the reindeer and sheep (Table 3). *Enoplastron* was found only in the sheep. All the ciliate genera showed large variation in their numbers, as also in earlier studies (WESTERLING 1970, SYRJÄLÄ, et al. 1973, SYRJÄLÄ et al. 1976). Some of the genera or subgenera were missing completely in some samples from both the reindeer and sheep.

The ciliates found in the rumen of the reindeer represented 15 different species. Of the 19 species found by WESTERLING (1970) and SYRJÄLÄ et al. (1973), the following were lacking in this study: *Ostracodinium confluens* and *Enoploplastron triloricatum,* here found in the sheep, and *Epidinium ecaudatum* and *Epidinium gigas.* The last-mentioned is specialized to the lichen feeding (WESTERLING 1970), which ecplains its absence in animals on a silage diet. *Entodinium damae* was the most common ciliate in this

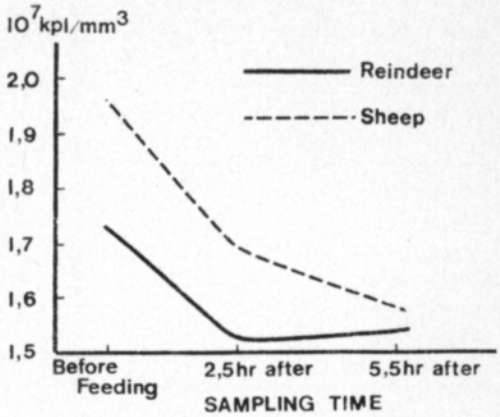

Fig. 3. The numbers of bacteria cells at the different sampling times.

experiment, as also in reindeer during the summertime (WESTERLING 1970). About 60 % of the numbers of *Entodinium* in the reindeer were made up by *E. famae*, *E. simplex* and *E. bicornutum*.

The different feeding was probably the main reason why the ciliate fauna on the reindeer in this experiment differed from those in the studies of WESTERLING (1970) and SYRJÄLÄ et al. (1973); in those earlier experiments the reindeer were mainly on a lichen diet. The descriptions of the ruminal fauna in the literature reveal large variations in the numbers and species composition, depending on nutritional and many other factors (HUNGATE 1966). In domestic ruminants on normal feeding, the fauna sometimes comprises more than 30 different species and sometimes less than 10.

Bacteria. The average number of bacteria was significantly lower in the reindeer than in the sheep ($P<0.01$, Table 3, Fig. 3). In the samples taken from reindeer before feeding, the number of bacteria was $17.3 \times 10^6/mm^3$, whereas the value for reindeer on a lichen diet was $6.7–9.7 \times 10^6$ mm^3 (SYRJÄLÄ et al. 1973). The corresponding value for the sheep in this experiment was $19.6 \times 10^6/mm^3$ and in an earlier experiment with silage feeding $35.0 \times 10^6/mm^3$ (SYRJÄLÄ et al. 1976).

Volume of the microbe mass

The volumes of both the ciliates and bacteria differed significantly ($P<0.001$ and $P<0.01$, respectively) between the reindeer and sheep (Tables 4–5). The total volume of ciliates was more than three times as high in the sheep as in the reindeer. In both animals the greatest contributions to the ciliate volume were made by the genera *Entodinium* and *Diplodinium*. In reindeer on a lichen diet the subgenera *Epidinium* made up 20–40 % of the ciliate mass (WESTERLING 1970, SYRJÄLÄ et al. 1973), whereas in this experiment it was lacking completely in the reindeer.

The proportion of the total volume of the microbe mass in the rumen contents averaged 1.8 % in the samples from the reindeer and 2.4 % in those from the sheep (Table 5). In the samples taken before feeding the corresponding values were 2.0 % and 3.0 %. These values are lower than in earlier studies of reindeer (WESTERLING 1970, SYRJÄLÄ et al. 1973) and sheep (SYRJÄLÄ et al. 1976).

Table 4. The volume of ciliates ($\mu m^3 \times 10^6$) per mm^3 rumen contents and its percentage distribution by genera and subgenera.

	Volume		Percentage distribution	
	Reindeer	Sheep	Reindeer	Sheep
Ciliates, total	2.3	7.3	100	100
Holotricha	0.1	0.5	4.3	6.9
Entodinium	0.7	2.9	30.4	39.7
Diplodinium	1.0	2.3	43.5	31.5
Eudiplodinium	0.3	0.6	13.1	8.2
Ostracodinium	0.2	0.9	8.7	12.3
Enoploplastron	0.0	0.1	0.0	1.4

Table 5. The proportion of the total microbe mass in the rumen contents and its distribution by ciliates and bacteria.

	Reindeer	Sheep
% of rumen contents:		
Ciliates	0.2	0.7
Bacteria	1.6	1.7
Total microbe mass	1.8	2.4
% of microbe mass:		
Ciliates	13	30
Bacteria	87	70

The proportion of bacteria in the reindeer microbe mass was 87 % and in the sheep 70 %. WARNER (1965) also reported that bacteria formed the greater part of the rumen microbial mass, whereas ABOU AKKADA (1965) found that the proportions of bacteria and ciliates are about equal in domestic ruminants.

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SELOSTUS

Säilörehun hyväksikäytön vertailu porolla ja lampaalla
2. Pötsikäyminen ja pötsin mikrobisto

Liisa Syrjälä-Qvist

Helsingin yliopiston kotieläintieteen laitos

Pötsin sisällön pH, ammoniakki ja haihtuvat rasvahapot sekä mikrobisto olivat kriteereinä verrattaessa säilörehun hyväksikäyttöä porolla ja lampaalla.

Pötsin käymistapahtumat olivat porolla vähäisemmät eivätkä kriteerit eri näytteenottoaikoina (ennen ruokintaa, 2½ ja 5½ tuntia ruokinnan aloittamisesta) poikenneet toisistaan niin paljon kuin lampaalla. Pötsinesteen pH oli porolla keskimäärin eri näytteenottoaikoina 6.94 ja lampaalla 6.61. Vastaavat ammoniakkimäärät olivat 17.0 ja 24.2 mg NH_3–N/100 ml sekä haihtuvien rasvahappojen kokonaismäärät 8.46 ja 10,90 mmol/100 ml. Etikkahapon osuus rasvahapoista oli porolla 75.3 mooli–% ja lampaalla 66.0 mooli–%. Vastaavat propionihapon mooliosuudet olivat 18.5 % ja 22.0 % sekä voihapon 4.2 % ja 8.8 %.

Alkueläimiä oli poron pötsin sisällössä keskimäärin 87 kpl/mm^3 ja lampaan 314 kpl/mm^3. Bakteerien määrät olivat vastaavasti 16.0×10^6 ja 17.4×10^6 kpl/mm^3. Mikrobimassan kokonaismäärä pötsin sisällöstä oli porolla 1.8 % ja lampaalla 2.4 %. Bakteerien osuus tästä mikrobimassasta oli porolla 87 % ja lampaalla 70 %. – Porojen ja lampaiden väliset erot pötsikäymisiä kuvaavissa kriteereissä ja mikrobimäärissä olivat lähes kaikissa tapauksissa tilastollisesti merkitseviä ($P<0.001$, $P<0.01$ tai $P<0.05$).