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# Dairy sheep and goats sort for particle size and protein in mixed rations



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# ABSTRACT

Sorting for specific parts or nutrients in feed is a natural behavior of ruminants, but for intensive dairy systems it is disadvantageous if it leads to nutritional imbalances for the individual or the herd. To prevent feed sorting in cattle, mixed rations (MR) are fed and forage components are cut as short as possible. In small ruminants, feed sorting is well documented but not well studied in relation to MR. The aim of this study was to investigate the ability of dairy sheep and goats to feed sort in MR. Experiments were conducted with each of 12 pairs of female adult dairy sheep and goats. In the first experiment, three MR composed of different forages (hay, grass silage, maize silage) were tested consecutively, each in a long and short cutting length variant. In the second experiment, two short cut variants of a grass silage-hay MR were investigated that differed in nutritional value. For all experiments, animals received each variant of MR for five consecutive days. The composition of feed rests at 11:00, 15:00 and the next day were compared to the ration fed at 09:00. Both species sorted all of the offered MR for large particles, and sheep but not goats sorted for protein. In the short cut MR variants, particle size sorting was reduced in the first two hours after feed delivery (09:00-11:00), but cutting length had no relevant effect thereafter, or on protein sorting. The nutritional value of the MR had no detectable influence on feed sorting. Both species sorted for larger particles in both variants, and goats sorted against protein in the first two hours after feed delivery, whereas sheep sorted for protein thereafter. These results show that sheep and goats are able to change the composition of MR within two hours after feed delivery. A short cutting length delayed feed sorting to a limited extent. Maintaining the feed quality of a mixed ration throughout the day is important for the health and welfare of dairy sheep and goats but seems to be a major challenge for feeding management.

## 1. Introduction

As part of their natural feeding behavior ruminants can be observed taking up feed selectively by sorting for specific plants or plant parts. In the wild, feed sorting serves as a strategy to meet the animal's nutritional demands with the plant species available for a balanced diet (Duncan et al., 2006), and it can also be observed under husbandry conditions, in which its extent also varies depending on the nutritional value of a ration (Madruga et al., 2017).

Ruminants sort between plant parts by particle size. For example, it has been shown that dairy cows sort against large particles (Maulfair et al., 2010). By doing so, they can change the nutritional value of their feed as different-sized plant parts also differ in their nutritional values. However, in intensive dairy production with rations of high nutritional value, sorting against large particles of high fiber content (Savadogo et al., 2000) and for small particles can lead to an excessive intake of highly fermentable carbohydrates (Ramanzin et al., 1986), that lower rumen pH (Miller-Cushon and DeVries, 2017). At the individual level, feed sorting should therefore be prevented under intensive feeding to ensure rumen health and to avoid undesirable changes in milk composition (Bhandari et al., 2008). At the herd level, feed sorting is problematic due to the feed competition between the animals. If the quality of the ration declines over time after feed delivery, animals that have access to the feed later (e.g. low-ranking animals) receive feed of an inadequate nutrient composition. The sorting behavior of cattle was found to be even stronger when competition for the feed is increased, for instance, by low feeding frequency or low amounts of feed per animal (Kronqvist et al., 2021; Sova et al., 2013). In the majority of these studies, feed sorting is evaluated by comparing the composition of the ration fed to that of leftovers 24 hours after feed delivery. However, to evaluate the time point at which the quality of the feed is substantially diminished, it would be necessary to investigate the extent of sorting throughout the day.

High-producing dairy animals need to consume a balanced ration

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Composition per kg DM of experimental mixed feed rations of Exp. 1 (HH = 1st and 2nd cut grass hay 50:50, GH = grass silage and grass hay 50:50, MG = corn, grass silage and alfalfa hay 40:55:5) and mixed ration variants of Exp. 2 (GH2 in variants A = low nutritional value and B = high nutritional value).

		Requir	ement/ kg intake/ animal <sup>1</sup>	Experim	ent 1		Experiment 2						
				нн		GH		MG		GH2 A		GH2 B	
Parameter		Goat	Sheep	mean	±SD	mean	±SD	mean	±SD	mean	±SD	mean	±SD
DM	kg			0.87	0.04	0.65	0.02	0.52	0.02	0.62	0.02	0.43	0.03
OM	g/kg			926	2.7	918	3.4	933	2.6	923	8.6	915	5.8
CP	g/kg	132	76	90.3	2.6	90.2	2.6	92.3	3.4	88.2	3.1	106	6.1
ADF	g/kg	190	190	277	8.5	301	13.5	257	12.3	269	10.7	257	11.1
NDF	g/kg	410	410	470	12.5	484	12.5	421	15.6	444	12.6	425	16.9
Calcium	g/kg			3.0	0.2	4.2	0.2	5.0	0.5	4.5	0.8	5.7	0.5
Kalium	g/kg			23.9	1.6	27.2	1.6	20.5	1.2	21.5	1.2	21.8	0.9
Magnesium	g/kg			1.4	0.1	1.6	0.1	1.7	0.0	1.7	0.2	1.9	0.1
Natrium	g/kg			0.1	0.0	0.1	0.0	0.1	0.1	0.1	0.1	0.2	0.1
Phosphor	g/kg			2.9	0.1	3.4	0.1	3.0	0.1	3.0	0.2	3.0	0.3
$NE_L^2$	MJ/kg	7.4	4.2	5.5	0.0	5.1	0.1	5.6	0.1	5.3	0.1	5.4	0.1
ADPE <sup>2, 3</sup>	g/kg	53	46	79.1	0.9	68.5	1.0	66.1	0.9	69.5	0.7	73.7	0.9
ADPN <sup>2, 3</sup>	g/kg			56.5	1.7	56.5	1.7	57.7	2.1	55.3	2.0	67.2	3.9

1) Requirements of CP, NE<sub>L</sub> and ADP(E/N) calculated according to Agroscope (2021); ADF and NDF recommended by Lu et al. (2005). with a mean BW of 67.9 kg and mean daily DM intake of 1.0 kg for goats and a mean BW of 78.9 kg and mean daily DM intake of 1.4 kg for sheep.

2) Calculated according to Agroscope (2021).

3) ADPE /N = Absorbable protein at the duodenum limited by rumen fermentable energy /nitrogen.

that meets their nutritional requirements. A common approach to reducing feed sorting and providing a balanced ration is feeding ruminants with mixed rations (MR), within which most or all feed components (forages, concentrates, minerals) are mixed together (Schingoethe, 2017). Several properties of MR can further reduce feed sorting (Miller-Cushon and DeVries, 2017). Reducing the particle size of the forage components produces a more homogenous ration (Suarez-Mena et al., 2013). However, particles need to be of a minimum length so that rumination is not reduced by an insufficient structure of fibers (Maulfair and Heinrichs, 2013). Another factor studied to reduce feed sorting in MR is the DM content, in which it is expected that in a wet MR finer particles (leaves) will adhere better to larger ones (stems). However, the results of this method are inconsistent among studies, showing increased (Felton and DeVries, 2010; Miller-Cushon and DeVries, 2009), decreased (Kronqvist et al., 2021; Leonardi et al., 2005) or no effect on feed sorting (Fish and DeVries, 2012).

In small ruminants, feed sorting is well documented. On pasture, sheep and goats choose plant species with seasonal variety, and considerable differences between these animals have been found (Animut and Goetsch, 2008; Mohammed et al., 2020; Osoro et al., 2013). Goats show a preference for shrub and tree species compared to sheep (which prefer grass species) (Pande et al., 2002), and goats are particularly good at selecting soft plant parts higher in nutritional value from stiffer, more lignified parts (Bartolomé et al., 1998; Celaya et al., 2007). In indoor feeding, both species sort in a variety of different types of forage feeds (hays and barley straw: Hadjigeorgiou et al., 2001; Wahed and Owen, 1986; Wahed et al., 1990; wheat straw: Biswal et al., 2000; oat hay: Dutta et al., 1999; green maize: Dutta et al., 2000; oat straw: Islam et al., 1997). When comparing species, goats have been found to sort more strongly for small particles than sheep (Hadjigeorgiou et al., 2001, 2003), and sheep have been found to sort more strongly for nitrogen (a proxy for protein) and against ADF in hay and barley straw rations (Wahed and Owen, 1986).

While these results are all based on single feeds, the literature studying the feed sorting of sheep and goats in MR is scarce, and the trend for MR feeding in these animals is increasing (Pulina et al., 2013). Due to their well-documented abilities to sort food, it can be predicted that small ruminants are also able to sort feed in MR, and this should occur relatively quickly after providing feed. The aim of this study therefore was to investigate the abilities of dairy sheep and goats of feed sorting for particle size and protein in MR composed of different forages (hay, grass silage and maize silage) throughout the day.

# 2. Materials and methods

It was hypothesised that long cutting and low/varying quality of the components would increase feed sorting, whereas short cutting and high/equal quality of the components would reduce it. In relation to cutting length, all rations were expected to provide sufficient mastication during feeding and rumination for healthy rumen functioning. We further predicted that feed sorting would be detectable within the first two hours after morning feeding but more pronounced in the leftovers of the next morning. Between the species, we expected to find more protein sorting in sheep than in goats and stronger particle size sorting in goats than in sheep.

All animal care and experimental procedures were performed in accordance with the relevant legislative and regulatory requirements and the ASAB/ABS Guidelines for the Use of Animals in Research (ASAB & ABS, 2020). The Cantonal Veterinary Office, Thurgau, Switzerland (Approval No. TG10/18–30902) approved all procedures involving animal handling and treatment.

# 2.1. Animals and housing conditions

Two experiments were conducted at the Agroscope Research Station in Ettenhausen, Switzerland. The sample size included 24 female dairy goats (10 Saanen, 11 Chamois Colored goats and 3 crossbreeds) and 24 female dairy sheep (20 Lacaune and 4 East Friesian sheep). Two sheep and two goats were replaced between the experiments, due to social compatibility. All animals were three years old and had never lactated or been pregnant. In the first experiment, the mean body weight of the goats was 67.9 (standard deviation  $\pm 8.3$ ) kg, and the mean body weight of the sheep was 77.7 ( $\pm 8.5$ ) kg. During the second experiment, the goats and sheep weighed, on average, 70.7 ( $\pm 7.9$ ) kg and 95.4 ( $\pm 9.7$ ) kg, respectively.

Prior to and between the experimental phases and during habituation phases (see below), the sheep and goats were kept in the same barn in an outdoor climate with three pens for each species. Groups of nine animals were kept per pen, with eight experimental animals and one substitute animal per pen. Each goat pen had a total area of 17.5 m<sup>2</sup> (4.5 m × 3.9 m). Each pen had one drinker for *ad libitum* access to water and one mineral supply. Feed troughs with a palisade feeding fence (35 cm and 40 cm feeding space per animal for goats and sheep, respectively) were placed along the entire long axis of each pen.

The experiments were conducted in a separate outdoor climate barn

consisting of four sheep and four goat pens, each large enough to house two animals (as described in Berthel et al. 2022). Each pen was 2.4 m  $\times$  3.5 m and included an elevated feeding area with two places equipped with a trough and *ad libitum* access to water. Both animal of a pair had access to both feeding places. The two feeding places were separated by a solid wood wall (1.4 m  $\times$  0.6 m) to minimize agonistic interactions (Aschwanden et al., 2009a), and allowed for visual contact in the area above the trough. The litter area was bedded with sawdust.

## 2.2. Experiment 1: cutting length

## 2.2.1. Experimental feeds

The animal pairs received three different MR consisting of forages fed *ad libitum*. The MR (Table 1) were offered consecutively at different periods in winter 2019/2020. The first MR consisted of first and second cut grass hay (HH; DM ratio 50:50), which typically differ in their chemical composition (see supplementary Table S1) and was tested in November 2019. The second MR consisted of grass silage and grass hay (GH; DM ratio 50:50) and was tested in January–February 2020, and the third MR consisted of corn silage, grass silage and alfalfa hay (MG; DM ratio 40:55:5), tested in March–April 2020. Each MR was offered in two cutting length variants, a long (6–8 cm) and a short (3–4 cm) variant. Each type of MR was habituated in the home pen for 14 days by switching the long and short variants daily.

# 2.2.2. Experimental procedure

The experiment was conducted with 24 sheep and 24 goats. As the access to the trough was not individual in the experimental pens (see Section 2.1) all feed data was collected on animal pairs. This resulted in experimental units of 24 pairs (12 pairs of sheep and 12 of goats) which were tested on both variants per experiment (schematic figure of habituation and experimental phases is shown in the supplementary figure S2).

The experimental phase of Experiment 1 consisted of 10 days for each type of MR in which both cutting length variants were presented (Berthel et al., 2022). The animals received the long variant (L) for five days and the short variant (S) for five days. The order of the variants was L to S for half of the pairs and S to L for the other half. Eight pairs were tested simultaneously, so that the procedure was repeated for three groups of eight pairs (four pairs of sheep and four pairs of goats, with two pairs of sheep and goats having the orders S to L and L to S). Animals were fed *ad libitum* with three feed deliveries a day (time points: 09:00, 11:00 and 16:00). At each feed delivery, all feed rests were removed from the troughs and weighed, representative samples were taken and fresh feed was provided. Animals received 28–33 %, 31–40 % and 30–40 % of their total daily feed at 09:00, 11:00 and 16:00, respectively. Total feed was provided at 140–150 % of daily intake to ensure ab libitum feeding at all times.

## 2.2.3. Data collection

The experimental procedure and data collection were the same for all three tested MR. The three MR were tested consecutively in the order of HH, GH and MG, with the habituation phases (14 days) and hay feeding in between. On the last two days of the five-day experimental period per variant, the feed delivered and the feed rests at each time point (11:00, 16:00 and 09:00 the next day) were weighed to calculate feed intake per pair by their differences. The arithmetic mean and standard deviation of the mean were calculated across pairs for each MR variant and divided by two as an estimated for individual intake. The intake (per pair) was additionally corrected to the live weight of the pair. The live weight of a pair was calculated by the sum of the two individual body weights (BW; in kg) to the power of 0.75 (BW<sup>0.75</sup>).

One feed sample was taken per day from each ration of the MR variants (S and L) directly after mixing before feeding. From the feed rests, feed samples were collected from each pair at each time point, resulting in six samples of feed rests per MR variant and pair. All samples

were analysed directly after sampling for particle size distribution using a Penn State Particle Separator (PSPS; See Section 2.2.1) and subsequently dried at 60 °C (HH: 24 h; all other MR: 48 h) to calculate DM content. The samples of the rations were pooled over the two days of data collection to perform a full wet chemical analysis (for exact methods, see Section 2.2.5). The samples of the feed rests at 11:00 were pooled over the two days of data collection per pair, resulting in one feed rest sample per variant and pair to analyse CP and crude ash (see Section 2.2.5).

## 2.2.4. Particle size analysis of feed samples

For analysis of particle size, we used the PSPS (Shaky 4.0, Wasserbauer, Austria), which consists of three screens with round holes of 19 mm, 8 mm and 4 mm in diameter and a bottom pen. For a standardized shaking of each of the 28 feed samples per day, the PSPS was placed on an electronic laboratory platform shaker (Agitateur Rotatest, Model 35|9B|0B, Lab-line Instruments, Inc., Melrose Park, Ill. 60160). A first shaking period of all sieve levels assembled was performed at 210 rotations per min for 60 s. After removing the top level, it was shaken again at 250 rotations per min for 20 s. After removing the second top level, the last sieve and pan were shaken again at 250 rotations per min for 20 s. In a pre-test, this method led to comparable results to the manual shaking method of the user manual of the Shaky 4.0.

With the proportional weight left on each level of the PSPS and the hole size of the screens, the mean particle size (mPS) was calculated for each sample according to ASABE Standard S424.1 (ASABE Standard, 2017). To calculate the mean length of particles for each level of the PSPS, the hole diameters of the screen above and the screen of the respective level were used. For the calculation of the top level, the hole diameter of the upper screen was replaced by the cutting length of the rations (80 mm for the long variants and 35 mm for the short variants).

## 2.2.5. Chemical analysis of feed samples

For the chemical analyses, the samples of the rations and the feed rests at 11:00 (and 09:00 the next day for Exp. 2), and the dried samples were ground to pass through a 1 mm screen (Brabender rotary mill; Brabender GmbH & Co. KG, Duisburg, Germany). They were analysed for exact dry mass content by heating at 105 °C for three hours (prepASH, Precisa Gravimetrics AG, Dietikon, Switzerland) and subsequently for ash content after incinerating at 550°C until a stable mass was reached according to ISO 5984\_2002. Organic matter was calculated by subtracting the ash content from the DM content. The CP content was calculated as the nitrogen content multiplied by a coefficient of 6.25, where N was determined using the Dumas method (ISO 16634-1:2008). The samples of the rations were also analysed for neutral detergent fiber (aNDF; ISO 16472:2006) and acid detergent fiber (ADF; ISO 13906:2008) contents with a fiber analyzer (Fibertherm Gerhardt FT-12, C. Gerhardt GmbH & Co. KG, Königswinter, Germany) and were expressed without residual ash. The NDF was analyzed with the addition of heat-stable amylase and sodium sulphite. Mineral content was analyzed according to EN 15510:2008 by ICP-OES (ICP-OES 5800, Agilent Technologies, Switzerland) after microwave digestion. Samples were dissolved in a glass tube (5 ml HNO3 65 % + 3 ml H<sub>2</sub>O ASTM Class I) using a microwave digester (UltraClave MLS, Leutkirch, Germany) at 235°C for 60 min (1000 W).

# 2.2.6. Monitoring feeding behavior

To control for sufficient and equal mastication on the short and long variants, all animals were equipped with MSR feeding behavior monitoring halters (JAM-R; Berthel et al., 2023; Nydegger et al., 2010) for 48 h on the last two days of the five-day experimental period per variant of all three MRs. The automatically recorded data by JAM-R were evaluated for the number of mastications while feeding (Mfeed) and while ruminating (Mrumi), and reported per day (by dividing the numbers of the 48 h recordings by two).

## 2.3. Experiment 2: Nutritional value

## 2.3.1. Experimental feeds

In the second experiment, conducted in April–May 2021, the animals received a mixed ration consisting of short cut (3–4 cm) grass hay and grass silage (GH2; DM ratio 50:50; Table 1). This ration was offered in two variants of differing nutritional value. They were prepared by mixing the same hay with grass silage of lower nutritional value and higher DM (variant A) and one of higher nutritional values and lower DM (variant B). This resulted in the two MR variants that differed in their chemical composition of DM, ADF, NDF and CP (all  $p \le 0.01$  based on linear models). The habituation phase for the MR lasted for 14 days, switching between the A and B variants daily.

## 2.3.2. Experimental procedure

In Experiment 2, again 24 animal pairs were tested like in experiment 1 (see Section 2.3.2). The experimental phase of experiment 2 also lasted 10 days with 5 days on either variant A and B and half of the pairs with alternating order of the variants. The feeding schedule in experiment 2 differed from experiment 1: Animals were fed *ad libitum* with one feed delivery per day (09:00) in which 120 % of the daily intake that had been eaten the day before was provided. After two hours (at 11:00), all feed rests were removed from the troughs, but after weighted and representative (220–340 g) samples were taken from it, the feed was relocated into the troughs. No further manipulations or additions to the feed were conducted.

#### 2.3.3. Data collection

In experiment 2, data collection also took place on the two last days of the five-day experimental period per variant. Feed intake was taken for the periods from 09:00–11:00 and 11:00 to the next day. Feed samples and samples of feed rests were taken and pooled the same way as in experiment 1 (see section 2.4.3), except that feed rests at 11:00 and the next morning were analysed for both PS and CP.

Analysing methods for PS and CP are described in sections 2.3.4 and 2.3.5.

Feeding behaviour was assed as described in Section 2.2.6.

# 2.4. Statistical analysis

For the statistical analyses and data visualisation, we used the opensource software R version 4.2.0 (R Core Team, 2021). To assess whether the data met the model assumptions QQ-plot and residuals versus predicted values-plot were inspected using the *simulateResiduals* function of

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the *DHARMa* R package (Hartig, 2017). As MRs were tested consecutively (not simultaneously) and were neither isoenergetic nor isoproteic, all comparisons were made between variants within each MR but not between MRs.

## 2.4.1. Feed sorting

To analyse sorting for particle size, changes in mPS after feed delivery were investigated as an outcome variable. To avoid the statistical limitations associated with compositional data, the change in mPS was expressed as a proportion of the mPS of the feed rests to the mPS of each respective MR ration variant, transformed by log ratio. When the proportion is zero, the mPS of the feed rests is the same as that of the ration fed. Negative values indicate that the mPS of the rests is lower than the mPS of the ration as fed. Positive values indicate that the mPS of the rests is higher than that of the ration as fed.

For each MR in Exp. 1 and the MR in Exp. 2, the change in mPS was analysed in a separate linear mixed-effect model using the *lmer* function of the *lme4* R package (Bates et al., 2015). The models included an intercept for each time point individually for both feed variants and both species as a fixed effect. The model formula is as follows:

Log (mPSrest/mPSration)  $\sim 0$  + Time: Feedvariant: Species + (1] Group/Pair) + (1] Date).

To analyse sorting for protein, the natural logarithm of the ratio of the CP of feed rests to the CP content of the MR as fed was calculated and analysed using linear mixed-effects models. In the case of Exp. 1, no effect for time needed to be included as CP was analysed at only one time point (11:00). For Exp. 2, the fixed effects and their three-way interaction of time, variant and species were included:

Log (CP<sub>rest</sub>/CP<sub>ration</sub>) ~ 0 + Feedvariant: Species + (1| Group/Pair) + (1| Date) (Exp. 1)

Log (CP<sub>rest</sub>/CP<sub>ration</sub>) ~ 0 + Time: Feedvariant: Species + (1| Group/Pair) + (1| Date) (Exp. 2)

In all models, a random intercept for pair nested within group (1) Group/Pair) accounted for repeated testing of the same animal pair over replicate days and for potential effects of group affiliation, and was included the crossed random effect of date (1) Date) for variance occurring by the day of data collection.

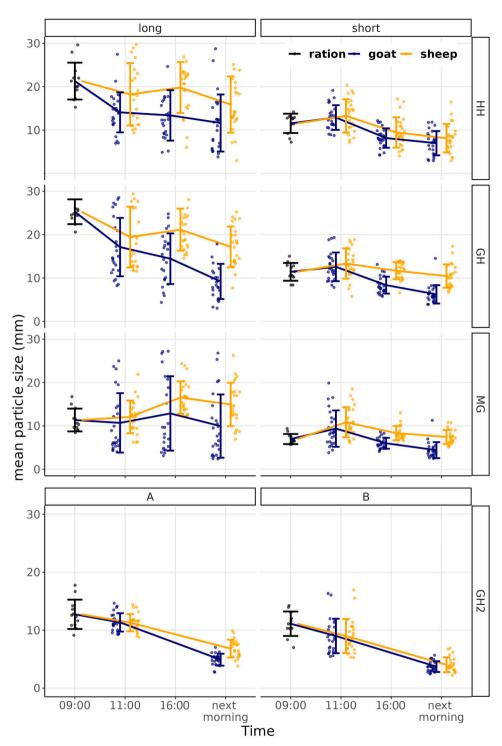
To assess the general impact of the fixed effects, the full model was tested against the null model without fixed effects and only random effects using the *anova* function (Bates et al., 1992).

The presence or absence of feed sorting in MR was assessed using bootstrapped 95 % quantile confidence intervals (CI<sub>95 %</sub>), which were

### Table 2

Mean ( $\pm$  standard deviation) individual DM intake and percentage of leftovers per day of all experimental MR rations (HH = 1st and 2nd cut grass hay, GH = grass silage and grass hay, MG = corn and grass silage; Table 1) and their variants during Exp. 1 (cutting length) and Exp. 2 (nutritional value: A = low nutritional value and R = high nutritional value)

B = high nutrition						et			
	Unit	Goats				Sheep			
Experiment 1									
HH		short		long		short		long	
DM intake	g	941	$\pm 218$	966	$\pm 128$	1416	$\pm 171$	1331	$\pm 215$
corrected	g/kg LW	41	$\pm 9$	42	$\pm 5$	57	±7	53	±9
leftover	%	54	$\pm 12$	53	$\pm 8$	38	±9	42	$\pm 11$
GH									
DM intake	g	935	$\pm 123$	975	$\pm 237$	1258	$\pm 182$	1299	±177
corrected	g/kg LW	40	$\pm 6$	42	$\pm 9$	50	$\pm 8$	52	±7
leftover	%	41	±7	41	$\pm 13$	43	$\pm 9$	40	±7
MG									
DM intake	g	1131	$\pm 261$	1225	$\pm 255$	1420	$\pm 228$	1400	$\pm 165$
corrected	g/kg LW	49	$\pm 11$	53	$\pm 11$	57	$\pm 10$	56	±7
leftover	%	39	$\pm 11$	35	$\pm 11$	40	$\pm 9$	41	±7
Experiment 2									
GH2		Α		В		Α		В	
DM intake	g	1043	$\pm140$	1038	$\pm 164$	1538	$\pm 196$	1589	$\pm 127$
corrected	g/kg LW	42	$\pm 6$	42	±7	52	$\pm 5$	54	$\pm 3$
leftover	%	21	$\pm 5$	17	±7	18	$\pm 6$	12	$\pm 3$



**Fig. 1.** Observed particle size of the mixed rations fed at 9:00 and of the feed rests at 11:00, 16:00 (only Exp. 1) and the next morning of sheep and goats in Exp. 1 (top panel, of the mixed rations HH = 1st and 2nd cut grass hay, GH = grass silage and grass hay, MG = corn and grass silage; in variants short and long) and Exp. 2 (bottom panel, of the mixed ration GH2; in variants A = low nutritional value and B = high nutritional value). Dots are single observed raw data values and lines connect mean values  $\pm$  SD with errorbars.

determined via parametric bootstrapping as implemented in *bootMer* (10,000 bootstraps, R package lme4). A significant difference from a null value (0) at the 0.05 level was assumed when the  $CI_{95\%}$  did not include the null value. Based on these calculations, feed sorting for particle size or protein was inferred to be present when the  $CI_{95\%}$  did not include zero. The direction of feed sorting can be derived from the range of the CI 95\% being positive or negative. A positive CI 95\% indicates sorting for small particles or against CP. A negative CI 95\% indicates sorting for

large particles or for CP.

To analyse the differences between the variants (S and L or GH2A and GH2B) and species, linear contrasts were set up using the *glht* function of the R package multcomp (Hothorn et al., 2008). A significant difference was assumed when the bootstrapped  $CI_{95~\%}$  (*bootMer*, 10,000 bootstraps) did not include the null value.

Mean content of crude protein (g/kg DM) of all experimental MR rations (HH = 1st and 2nd cut grass hay, GH = grass silage and grass hay, MG = corn and grass silage; Table 1) and their variants during Exp. 1 (cutting length) and Exp. 2 (nutritional value: A = low nutritional value and B = high nutritional value).

	Experiment 1	L		Experiment 2						
	НН		GH		MG		GH2			
	short	long	short	long	short	long	A	В		
Ration										
at time of feeding 09:00	100.5	97.4	97.5	97.2	98.3	101.4	88.3	106.7		
Rests										
Goats										
at 11:00	99.9	93.7	99.0	95.9	96.5	96.1	95.7	112.0		
the next morning							96.9	113.6		
Sheep										
at 11:00	92.2	86.6	94.7	89.7	95.0	94.6	91.9	106.5		
the next morning							85.3	101.8		

# 2.4.2. Feeding behaviour

To analyse feeding behaviour, Mfeed and Mrumi were investigated as outcome variables. For both outcome variables and each of the three MR in Exp. 1 and the MR in Exp. 2, a linear mixed-effect model using the *lmer* function of the *lme4* R package (Bates et al., 2015) estimated the effect of species, MR variant and their interaction. The model formula is as follows:

Mfeed or Mrumi ~ Feedvariant: Species + (1| Group/Pair) + (1| Date)

In all models, a random intercept for pair nested within group (1 | Group/Pair) accounted for repeated testing of the same animal pair over replicate days and for the potential effects of group affiliation, and included a crossed random effect of date (1| Date).

To assess the general impact of the fixed effects, the full model was tested against the null model without fixed effects and only random effects using the *anova* function (Bates et al., 1992).  $CI_{95~\%}$  were determined via parametric bootstrapping as implemented in *bootMer* (10,000 bootstraps). A significant difference between the levels of the fixed effects (variant, species) was assumed when the  $CI_{95~\%}$  did not overlap.

## 3. Results

# 3.1. Descriptive statistics

Feed intake and its variation between animals and days for all the MR and variants are listed in Table 2. The calculated mean particle sizes (mPS) of all tested MRs and feed rests at all measured time points for sheep and goats are shown in Fig. 1. The calculated mean CP of all tested MRs and feed rests at the measured time points are presented in Table 3.

## 3.2. Particle size sorting

For all models analysing particle size sorting, the global model comparison yielded statistically supported differences (HH/GH/MG Exp. 1 and GH2 Exp. 2: all p < 0.01). In Experiment 1, particle size sorting was detectable in all rations but not in every time period (Fig. 2, Supplementary Table S3). In HH and GH, goats sorted for large particles in both variants at all time periods except the short version in the first period (09:00–11:00). Sheep sorted in HH both variants in the latest period (16:00 to the next morning). In GH, they did not sort the short variant, but they sorted the long variant in all periods. MG was sorted less, with goats sorting for large particles in both variants from 16:00 to the next morning, and sheep sorting for small particles in the short variant from 09:00–11:00 and in the long one from 11:00–16:00.

In GH, the long variant was sorted more strongly than the short one by both species and at all periods Table 4. In HH and MG, this was the case only in the first period (09:00–11:00) for both species. Sheep sorted more strongly in the short variant of MG than in the long variant from 11:00–16:00. Overall, goats sorted more strongly than sheep, only in the short variant of HH, and no difference between the species could be detected.

In Experiment 2, during the period from 09:00–11:00, both species sorted for large particles in variant B but not A (Fig. 2). From 11:00 to the next morning, both species sorted both variants. No difference in sorting were observed for goats between the A and B variants. Sheep sorted B more strongly than A in the period from 11:00 to the next morning Table 4. Goats sorted more strongly than sheep for A, but no difference between species was found for B.

# 3.3. Sorting for crude protein

For all models analysing protein sorting, the global model comparison yielded statistically supported differences in all models (HH/GH/MG Exp. 1 and GH2 Exp. 2: all p < 0.001).

In Experiment 1, sheep sorted for CP in the long variants of HH, GH and MG as well as in the short variant of HH. Goats sorted for CP only in the long variant of MG (Fig. 3). For MG, both species sorted the long variant more strongly than the short one (Table 5). Sheep also sorted the long variant more strongly than the short variant in GH. For HH, no difference in sorting was found between the variants. Sorting for CP was stronger for sheep than goats in HH and GH but not MG.

In Experiment 2, goats sorted against CP in both variants and in both periods (Fig. 3). Sheep did the same for variant A from 09:00–11:00. From 11:00 to the next morning, they sorted for CP in both variants. No difference in CP sorting was found between the variants. Sheep sorted more strongly for CP in both variants and periods than goats, except from 09:00–11:00 of variant A (Table 5).

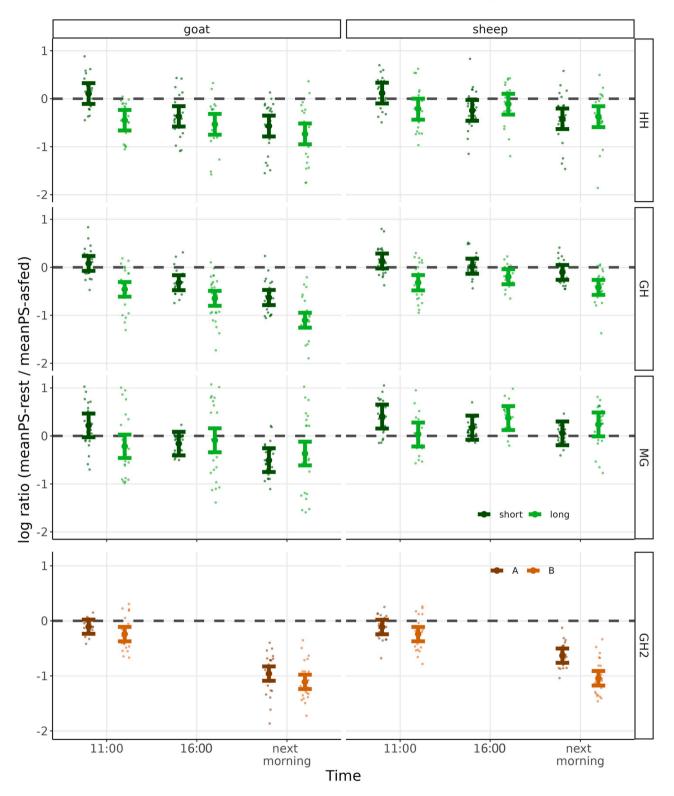
# 3.4. Feeding behavior monitoring

For all models analysing the number of mastications, the global model comparison yielded statistically supported differences for Mrumi (HH: p < 0.001; GH: p < 0.001; MG: p < 0.001) but not for Mfeed (HH: p = 0.25; GH: p = 0.74; MG: p = 0.71).

Mfeed and Mrumi were on similar levels throughout all rations of Experiments 1 and 2 (Supplementary Table S4). The estimated means for Mfeed per day across all rations ranged from 14,936 to 18,039 for goats and from 15,807 to 19,189 for sheep. The estimated means for Mrumi per day across all rations ranged from 22,757 to 26,549 for goats and from 33,828 to 37,258 for sheep. Within each ration, no differences were found between variants in Mrumi. The Mrumi per day was higher for sheep than goats in each MR (HH +9260, GH +9850, MG +8410, GH2 +13,120).

# 4. Discussion

In this study, we investigated the sorting abilities of sheep and goats in MRs consisting of different types of forage. Both species were able to sort these MRs for protein and particle size. According to our hypothesis,



**Fig. 2.** Sorting for particle size in sheep and goats. Estimated mean with bootstrapped 95 % confidence intervals (errorbars) of the (log transformed) ratio of the mean particle size (mPS) of feed rests to the mPS of rations fed in Exp. 1 (three top panels, of the mixed rations HH = 1st and 2nd cut grass hay, GH =grass silage and grass hay, MG = corn and grass silage; in variants short and long) and Exp. 2 (bottom panel, of the mixed ration GH2; in variants A =low nutritional value and B =high nutritional value) measured at 11:00, 15:00 and the next morning. The inclusion of the dashed line at y = 0 in a confidence interval represents no feed sorting; positive and negative values display sorting for small and large particles, respectively.

goats showed stronger particle sorting than sheep, and sheep sorted stronger for protein than goats. On the other hand, silage of lower quality was not sorted more than that of higher quality. The animals were able to sort all feeds within two hours of feed delivery, and cutting the MR feeds short could reduce sorting for particle size to some extent but did not influence CP sorting.

Sheep and goats were able to sort between different particle sizes in all the MR offered. Surprisingly, they generally sorted for large and

Bootstrapped 95 % confidence intervals (CI) of linear contrasts and an indication of the contrast hypothesis outcome (Hc) for particle sorting between variants and species for the mixed rations of Exp. 1 (HH = 1st and 2nd cut grass hay, GH = grass silage and grass hay, MG = corn and grass silage; Table 1) and Exp. 2 (GH2; in variants A = low nutritional value and B = high nutritional value).

	Experimer	nt 1								Experime	ent 2	
	НН			GH			MG			GH2		
	CI 95 % (f	rom; to)	Hc	CI 95 % (from; to) Hc			CI 95 % (	from; to)	Hc	CI 95 % (from; to)		Hc
Contrast hypothesis:		variant long - variant sho		$\operatorname{ort} = 0$					variant A - variant B =		= 0	
Goats												
09:00-11:00	-0.77;	-0.34		-0.71;	-0.37	_	-0.63;	-0.24	_	0.00;	0.26	0
11:00-16:00	-0.38;	0.04	0	-0.50;	-0.15	-	-0.12;	0.26	0			
16:00 to next morning	-0.37;	0.04	0	-0.64;	-0.30	_	-0.05;	0.33	0			
11:00 to next morning			Ū							0.02;	0.28	0
Sheep												
09:00-11:00	-0.54;	-0.12	_	-0.63;	-0.28	_	-0.56;	-0.18	_	0.00;	0.26	0
11:00-16:00	-0.08;	0.34	0	-0.39;	-0.05	-	0.01;	0.39	+			
16:00 to next morning	-0.17;	0.25	0	-0.49;	-0.14	-	-0.01;	0.38	0			
11:00 to next morning										0.28;	0.54	+
Contrast hypothesis:	goa	ts - sheep =	= 0									
Long Mean over periods	-1.53	-0.50	_	-1.72	-0.82	-	-2.06	-0.59	_			
Short A Mean over periods	-0.80	0.23	0	-1.37	-0.47	-	-1.83	-0.33	-	-0.56	-0.08	_
В										-0.30	0.17	0

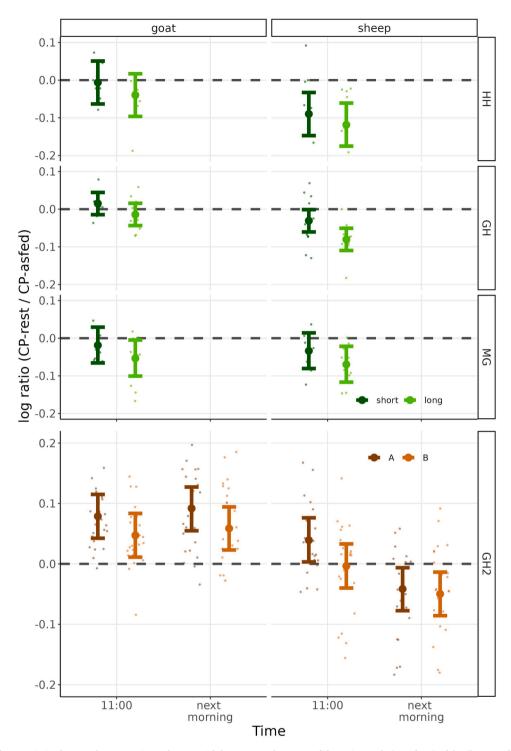
against small particles, in contrast to previous studies finding sorting for smaller particles in cattle (Miller-Cushon and DeVries, 2017) as well as sheep and goats (Hadjigeorgiou et al., 2001). Sorting for large particles has been observed in cows (DeVries et al., 2008) and goats (Giger-Reverdin, 2018) in response to a rumen acidosis challenge. The presence of rumen acidosis is unlikely in the present study, as no concentrate was offered in any of the experimental MR and ruminating behaviour was not detectably affected by cutting lenght. Concentrate components are rather small particles and are highly preferred by cattle (Miller-Cushon and DeVries, 2017) and sheep (Helander et al., 2014). Concentrate was fed in all MR in the above-reported studies and might explain why sorting for small particles was found. In the present study, animals could only sort between plant parts and different types of forages in the MR. Although we could not further specify them, our study demonstrates that even in MR, sheep and goats are capable of selecting specific particles effectively.

Both species also showed sorting in relation to protein. Sheep sorted for protein in both experiments. Goats did not sort for protein in Exp. 1 and sorted against it in Exp. 2. This is in accordance to previous studies showing that sorting for protein is stronger in sheep than in goats (Hadjigeorgiou et al., 2003; Wahed and Owen, 1986). Possibly influenced by their wool production (Cao et al., 2021), sheep have a higher demand for protein than (non-fibre-producing) goats (Salah et al., 2014). However, the nutritional values and amounts of the offered rations were calculated to meet the maintenance requirements of goats and sheep according to Agroscope (2021), making specific protein sorting unnecessary for maintenance at least for sheep. It is possible that with the breeding for wool production, an innate behavior for selective feeding for protein might have unintentionally been bred along with it. As the genetic traits for wool production persist in dairy sheep breeds like the ones used here (Lacaune and East Friesian), the innate behavior traits for selective protein feeding might also still be present. Dairy goat breeds like the ones used here (Saanen and Chamois Coloured) do not

originate from fibre-producing goat breeds. This could explain why the sheep in the present study were searching for CP in the MR offered, whereas goats did not, and this is in line with a study finding that fibre-producing goats did not differ from sheep in nitrogen sorting (Hadjigeorgiou et al., 2001). Further studies are needed to investigate whether sorting for protein-rich feed components in small ruminants is driven by their protein requirements or whether it is an innate behavior connected to fibre production.

Experiment 1 aimed to identify whether cutting length could reduce feed sorting in MR. Within the first two hours of morning feeding, particle sorting was lower in short than long MR variants, but in later periods of the day, the sorting was at the same level. Similarly, in a previous study, the cutting length of grass hay had little to no influence on the feed sorting of goats and sheep over 24 hours (Hadjigeorgiou et al., 2003). Analysing feed sorting during different periods of the day in the present experiment showed that a short cutting length could at least delay sorting after feed delivery. Cutting the feeds even shorter might further delay sorting, but it is unclear whether this would be negative for rumen health (Tafaj et al., 2007). The cutting length of 3-4 cm in the present study still produced normal feeding behavior, with the number of mastications not differing from the long variants. These numbers are also comparable to reported mastications of grass hay and grass seed straw, which were only slightly higher with the higher DM, ADF and NDF contents of these feeds (Jalali et al., 2012).

Experiment 2 investigated whether differences in feed quality in the MR variants of hay and grass silage, produced by varying DM content and the chemical composition of the grass silage, affected feed sorting. This hypothesis was based on studies in cows in which feed sorting was stronger in MR with forages of lower quality (Madruga et al., 2017). The nutritional value of A, with lower CP and higher ADF and NDF contents, was distinctly lower than that of B, so that the animals would have to select A more than B to obtain the same nutritional content per unit fed. Variant A was also drier than B, which could have facilitated sorting



**Fig. 3.** Feed sorting for protein in sheep and goats. Estimated mean with bootstrapped 95 % confidence intervals (errorbars) of the (log transformed) ratio of the CP content of feed rests to the CP of rations fed in Exp. 1 (three top panels, of the mixed rations HH = 1st and 2nd cut grass hay, GH =grass silage and grass hay, MG = corn and grass silage; in variants short and long) and Exp. 2 (bottom panel, of the mixed ration GH2; in variants A = low nutritional value and B = high nutritional value) measured at 11:00 (Exp. 1 and 2) and the next morning (Exp. 2). The inclusion of the dashed line at y = 0 in a confidence interval represents no protein sorting; positive and negative values display sorting for and against CP, respectively.

(Kronqvist et al., 2021; Leonardi et al., 2005) as particles were presented as looser in B than in A. However, we did not find significant differences in the level of feed sorting between the two variants. Both variants were sorted for particle size in a similar pattern to the short variant of GH in Exp. 1 (being the most comparable ration to GH2, although a direct comparison is not possible as the MRs were tested consecutively and were neither isoenergetic nor isoproteic). Hadjigeorgiou et al. (2001) demonstrated that sheep and goats did not sort differently for hays and straws that differed naturally or artificially in nutritional value. However, it is still possible that the animals sorted for further characteristics not measured in that study or ours. Further, the difference in quality between the rations might not have been strong enough to produce a distinct effect on sorting. It would be worth investigating which specific parameters of feed quality control feed sorting in relation to the animals'

Bootstrapped 95 % confidence intervals (CI) of linear contrasts and an indication of the contrast hypothesis outcome (Hc) of sorting for CP between variants and species for the mixed rations of Exp. 1 (HH = 1st and 2nd cut grass hay, GH = grass silage and grass hay, MG = corn and grass silage; Table 1) and Exp. 2 (GH2; in variants A = low nutritional value and B = high nutritional value).

Experiment 1													Experiment 2				
	HH CI 95 % (from; to)		GH Hc CI 95				MG CI 95 % (from; to)		Нс	GH2	CI 95 %	(from; to)	Hc				
Contrast hypothesis: var	iant long –	variant s	hort =	0								A – B = 0	)				
Goats:												Goats:					
09:00-11:00	-0.07;	0.00	0		-0.06;	0.00	0		-0.06;	-0.01	-		-0.01;	0.07	0		
11:00 to next morning													-0.01;	0.07	0		
Sheep:												Sheep:					
09:00-11:00	-0.07;	0.01	0		-0.08;	-0.02	_		-0.06;	-0.01	_		0.00;	0.09	0		
11:00 to next morning													-0.03;	0.05	0		
Contrast hypothesis: goa	ts – sheep	= 0										Goats - s	heep = 0				
Long:												A:	•				
09:00-11:00	0.03;	0.13	+		0.03;	0.10	+		-0.01;	0.04	0		0.00;	0.08	0		
11:00 to next morning													0.09;	0.18	+		
Short:												B:	,				
09:00-11:00	0.03;	0.13	+		0.01;	0.08	+		-0.01;	0.04	0		0.01;	0.09	+		
11:00 to next morning	- ,				,				,				0.07;	0.15	+		

#### nutritional requirements.

Previously, the feed sorting abilities of sheep and goats have only been studied using 24 h feed leftovers of forage feeds (Biswal et al., 2000; Dutta et al., 1999, 2000; Wahed and Owen, 1986; Wahed et al., 1990). If small ruminants change the composition of MR within only two hours, feed quality declines rapidly. In intensive housing of dairy animals where feed is often restricted in time and/or space, this decline in quality could intensify the feed competition between animals. It has been shown that feed sorting in cows is even increased in competitive feeding situations (Krongvist et al., 2021; Leonardi and Armentano, 2007) and can be reduced in sheep and goats with a higher allowance (Biswal et al., 2000; Dutta et al., 1999, 2000; Islam et al., 1997; Osafo et al., 1997). Lower production levels of goats of low social ranks compared to higher-ranking ones (Barroso et al., 2000) could be explained by the limited access to non-sorted feed. Thus, in addition to providing adequate rations, the importance of the feeding schedule (DeVries et al., 2005, Crossley et al., 2018), the available feeding space (Loretz et al., 2004; Aschwanden et al., 2008) and the design of the feeding place (Aschwanden et al., 2009a, 2009b; Nordmann et al., 2011), are emphasised to ensure access to feed of equal quality for all animals.

Our study was conducted in non-lactating and non-pregnant animals fed in pairs and with access to a feed allowance far beyond their intake. Although this limits the external validity of our results, we can assume that also under farming conditions sheep and goats will sort efficiently in mixed rations within a short period of time. As we also know, that sheep and goats prefer the single components to the mixed ration (Berthel et al., 2022), it is questionable, whether feeding mixed rations is the best option for feeding a balanced ration to small ruminants. Probably, the feeding management should rather be based on the animals' "nutritional wisdom" (Provenza, 1995; Provenza, 2003). This concept had been built on the evidence, that small ruminants select their diet efficiently to their individual and temporary needs (Fedele et al., 2002, Görgülü et al., 1996). It has also been concluded that engaging in selective feeding is beneficial to the health and welfare of sheep and goats (Villalba et al., 2010; Provenza et al., 2007).

## 5. Conclusion

Dairy sheep and goats were able to sort for particle size and protein in a variety of mixed rations composed of different forages. However, varying the nutritional value (CP, NDF and ADF) of a mixed ration of hay and grass silage did not produce differences in feed sorting and highlights the need to better understand which factors in a mixed ration control feed sorting. Sheep and goats were able to change the composition of a mixed ration within two hours of feed delivery. A short cutting length of the mixed rations delayed feed sorting to a limited extent. Maintaining the feed quality throughout the day is important for the health and welfare of dairy sheep and goats. As feed sorting cannot be prevented by mixed rations this seems to be a major challenge for the feeding management of small ruminants.

# CRediT authorship contribution statement

Nina Maria Keil: Conceptualization, Funding acquisition, Supervision, Writing – review & editing. Frigga Dohme-Meier: Conceptualization, Resources, Writing – review & editing. Roxanne Magali Berthel: Data curation, Formal analysis, Methodology, Visualization, Writing – original draft.

## **Declaration of Competing Interest**

We confirm that this work is original and has not been published elsewhere, nor is it currently under consideration for publication elsewhere. We have no conflicts of interest to disclose.

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# Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.applanim.2023.106144.

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