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DETERMINATION OF FAECES PARTICLE PROPORTIONS AS A TOOL FOR THE EVALUATION OF THE INFLUENCE OF FEEDING STRATEGIES ON FIBRE DIGESTION IN DAIRY COWS

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ABSTRACT

Aim of this study was to test the influence of feeding strategy on faecal particle size distribution as indicator of fibre digestion under on-farm conditions. Seventy-six cows from 11 farms, located in Switzerland and southern Germany were included. Cows were monitored up to eight times during one full lactation. All valid individual test days resulted in a total of 347 datasets. Milk yield and composition were recorded. Diet composition was described in a binary yes/no-indication for the presence or non-presence of different roughage components (fresh grass, grass silage, hay, maize silage and straw) and the exact amount of concentrates. Faecal samples were taken at each test day and sieved with a set of four wire-mesh screens with pore sizes of 4mm, 2mm, 1mm, and 0.3mm under running tap water. Dry matter proportions were calculated. The inclusion of maize silage and an increasing level of concentrates significantly increased the presence of faecal particles in the sieves. Maize silage increased the larger particles, while concentrates enhanced the smaller particle fractions. No effect of fresh grass and grass silage was found. Based on the results, the faecal particle fractions appear to be a practicable tool to predict influences of feeding systems on fibre utilization in dairy cows.

Key words: Dairy cattle, feeding system, forage, faeces, fibre utilization.

INTRODUCTION

Ruminants are extraordinarily efficient in the breakdown of fibre particles during rumination and digestion (Clauss *et al*., 2010), in particular grazers like cattle (Hummel *et al*., 2008). Along the gastrointestinal tract, the fibrous particle size is decreased from several centimetres to average diameters of 0.5 mm and less (Kovacs *et al*., 1997; Clauss *et al*., 2010). The far largest part of particle size reduction happens by chewing during ingestion and rumination (McLeod and Minson, 1988). A highly developed ability to sort smaller from larger particles in the fore stomach (Clauss *et al*., 2010; Bayat *et al*., 2011), in order to proceed the former to the small intestine while processing the latter by further mastication and ruminal fermentation, is an important factor for the high efficiency in fibre digestion by ruminants. However, the amounts of potentially digestible fibre, which are excreted with faeces, are considerable. For example, Bayat *et al*. (2011) found that Airshire cows, receiving grass silage and 8 kg of concentrates, excreted daily 1.5 - 2.0 kg of digestible NDF. This implies that there is a certain potential to optimise the fibre digestion, which is of particular significance, if the fibre utilization is regarded as an important contribution of ruminant livestock to a global sustainable resource management (O'Mara, 2012). This perspective puts the grazer-type ruminant (Van Soest, 1992) into its native ecological role: the digestion of fibrous plants, thus utilisation of rangelands and grasslands, rather than of concentrates, which are grown on arable land. In feeding systems, which base on the principle of maximising the input of forages and minimising the use of concentrates, increasing the efficiency of fibre utilization by livestock ruminants is a crucial issue.

Concerning nutrient digestion and utilization of feedstuffs, evaluations are usually based on standard feeding values and equations, which are developed on the national levels. However, these standards, which are developed in agricultural systems basing on high usage of concentrate supplements (e.g. NRC, 2001 or GfE, 2001), might be not optimal if feeding strategies are followed, which focus on maximal utilization of fibrous forages instead of grains (Ivemeyer *et al*., 2014). If the aim is to develop feeding evaluation and planning systems for concentrate-poor diets, additional criteria and solutions have to be developed. Evaluation systems are needed, which do not base on the possibility to counterbalance any nutrient imbalance by the supplementation with concentrates. In order to describe the feeding situation and its efficiency, practicable scoring systems for the available forages are needed (Spengler Neff *et al*., 2012), and can be complemented by cow parameters, such as animal health and performance, but additionally also by variables which describe intake behaviour and digestion efficiency. A parameter for the latter might be the faeces particles (Clauss *et al*., 2010; Kornfelt *et al*., 2013).

The aim of the current study was to investigate under on-farm conditions the evaluation of faeces particle size distribution as a measure for the digestion of fibrous plants. This should - as an indirect parameter - enable to estimate the influence of actual feeding strategies on fibre digestion under practice conditions, provided that roughage utilization is set as the main qualifying target and low or zero concentrate inputs are intended.

MATERIALS AND METHODS

Farms and animals: The study was conducted on 11 different dairy farms in southern Germany and in Switzerland representing a broad spectrum of feeding and production intensity. The farms represented either a high production level (> 9'000 kg milk/cow/year on average; three farms with Holstein cows, one farm with Brown Swiss), an intermediate production level (7'000 – 8'999 kg; two farms with Holstein cows, one farm with Swiss Red Pied) or a low production level (5'000-6'999 kg; three farms with Swiss Red Pied cows, one farm with Holstein). From each farm six to nine cows were observed, which had to fulfil the criteria that they already passed at least the first lactation, with an individual performance matching the predefined production level of the farm. Farms were situated in Switzerland or in southern Germany. Roughly half of the farms produced according to the standards of organic agriculture, however, the considered system descriptor was the milk yield level rather than the production system. In sum, 76 cows were included in the study. Each individual cow was monitored during a full lactation if possible, always at the official milk control test days of the respective breeding associations (once a month). Not all cows finished the whole lactation, but all completed test days were included in the data. Only individual test days with missing values were excluded. This resulted in a total of 347 valid datasets (test days of individual cows containing all data as defined below).

Sampling and analyses: On the test days, the actual daily milk yield was recorded and milk composition (contents of fat, protein, and urea) was determined by the local breeding associations with the standard procedures (near-infrared spectroscopy). Additionally all milk samples were analysed for acetone by a fluorimetric flow-injection method. At each sampling, the actual feeding for each individual cow was characterized by documentation of the presence of fresh green forage, grass silage, hay, maize silage and straw (in a binary yes/no- notation) and of the individually provided amount of concentrates per day.

Further, at each test day, at least 200g of fresh faeces was collected from each individual cow. The faecal samples were frozen at – 20°C and later thawed and sieved. The sieving method was adapted from Bae *et al*. (1981). The apparatus used (VWR International, Dietikon, Switzerland) consisted of four wire-mesh screens with a diameter of 20 cm which were arranged vertically. The top screen had a pore size of 4 mm, the second of 2 mm, the third of 1 mm and the lowest of 0.3 mm. A 100-g aliquot of the faeces fresh matter was solved in 500 ml water and subsequently put on the top screen. A jet of tap water washed the particles through the following screens. After 10-15 seconds the top screen was removed, and after another 10-15 seconds also the 2-mm screen was taken away. The third and fourth screens were washed until the outflowing water was clear. Subsequently the residues from the screens were dried for 12 hours at 105°C and reweighed. The same was done with a 50 g aliquot of the original sample to determine dry matter (DM) of the base material.

Statistical methods: Two different models for the analysis of variance (ANOVA) were calculated in order to estimate the main effects of different factors on faecal particle proportions in more detail. Because of confoundings, e.g. between feeding descriptors and farm, it was not possible to include all factors in one model; therefore, two models were chosen as two different perspectives on the matter. Both models were applied for all particle fractions in relation to total faecal DM mass and in relation to total particle mass (DM).

The first model considered the effects of farm, animal, lactation number, milk yield and sampling number (as a measure for stage of lactation). Here, a mixed model was applied as follows:

$$Y = \mu + F_i + LN_j + A_{ijk}(F_i{*}LN_j) + SN_l + MY + \varepsilon_{ijklm} \quad (1)$$

where F was the farm, LN was the lactation number, A was the animal within farm and lactation number, SN was the sampling number, MY was the milk yield and ε was the residual error of the model.

The second model was set up in order to estimate the effects of the forage characteristics and concentrate amounts. Here, farm was not included, because it was characterized by the feeding and thus strongly confounded with these factors. The same was true for animal. The general linear model applied was:

$$Y = \mu + P_i + H_j + GS_k + MS_l + S_m + CON_n + \varepsilon_{ijklmno} \quad (2)$$

where P was fresh pasture grass (yes or no), H was hay (yes or no), GS was grass silage (yes or no), MS was maize silage (yes or no), S was straw (yes or no), CON was the level of concentrates and ε was the residual error of the model. Both procedures included a type III analysis of variance.

Pearson correlations were calculated for the faeces particle fractions with the daily concentrate amounts and indicative milk characteristics, which were the acetone and urea content and the fat-protein ratio.

For a better illustration of the correlations, a principal component analysis (PCA) was carried out in order to reveal an overview over the main interdependences of feeding, milk related parameters and faeces characteristics of the whole dataset. No rotation was applied to the principal components. The two principal components with the highest Eigen-value (always > 1) were plotted in a two-dimensional model. The graph presented is based on the loading vectors, displaying the variables based on their respective value within the two principal components.

All statistical calculations were carried out with IBM SPSS STAT 21.

RESULTS

In the first ANOVA model, based on farm, cow and sampling date as main characteristics, neither cow nor any of the terms characterizing the individual animal (lactation number and sample number as a measure for stage of lactation) had an influence on the proportions of different particle fractions in total faeces. Only milk yield had partly a significant effect, being positively correlated with the fractions of 1-2 and >4mm. By contrast, the factor ‘farm’ had a highly significant influence on all particle fractions (Table 1).

There was a positive correlation between concentrate level and all faecal particle proportions (Table 2). Considering the second ANOVA model, where the impacts of dietary components were tested, the presence of maize in the diet and the level of concentrates were the main factors determining the proportion of particle fractions within total faecal DM (Table 3). The significance of concentrates was higher for the smaller fractions, while maize silage influenced the larger fractions of faecal particles. The presence of straw in the diet significantly increased the faecal fraction of 4mm. A group of milk characteristics, which are indicative for the cows’ metabolic status, were tested for their correlation with the faeces particle proportions. Here, milk urea concentration appeared in a slight but significant negative correlation with all faecal particle fractions (Table 4).

In agreement with the results from the ANOVA, the principal component analysis showed a very close relationship between all particle fractions in the faeces. These were also closely related to silages, concentrates, straw, and milk yield, but in clear opposition to hay and green forage (Fig. 1). The milk characteristics and the cows’ characteristics (lactation number and days in milk) were not clearly related to the axis between feed characteristics and faeces particles.

Table 1 Significance of cow, lactation number, farm, sample number and milk yield for the proportions of faeces particle fractions in total faecal dry mass

Faecal particle fractions	Cow (within farm*lactation no.)	Lactation no.	Farm	Sample no.	Milk yield
	***P*-value**	***P*-value**	***P*-value**	***P*-value**	***P*-value**
Sum of all fractions	0.709	0.309	<0.001	0.190	0.202
>0.3 mm – 1 mm	0.794	0.468	0.004	0.169	0.308
>1mm – 2mm	0.172	0.270	<0.001	0.208	0.028
>2mm – 4mm	0.136	0.436	<0.001	0.231	0.148
>4mm	0.487	0.782	<0.001	0.253	0.049

Table 2. Pearson correlations of the dietary level of concentrates (kg/d) with proportions of faeces particle fractions (g/kg DM).

Faecal particle fractions	coefficient	*P*-value
Sum of all fractions	0.626	<0.001
>0.3 mm – 1 mm	0.539	<0.001
>1mm – 2mm	0.472	<0.001
>2mm – 4mm	0.494	<0.001
>4mm	0.355	<0.001

Table 3. Proportion of faeces particles (g/100g [DM] faecal mass) depending on the dietary presence of various roughage components and the level of concentrates.

Faecal particle fractions	Forage component	Presence of component[1]		S.E.	*P*-value
		yes	no		
Sum of all fractions					
	Concentrate amounts				<0.001
	Fresh grass (pasture)	34.8	35.2	1.06	0.635
	Hay	35.7	34.2	1.07	0.093
	Grass silage	35.5	34.4	1.18	0.285
	Maize silage	38.2	31.7	1.28	<0.001
	Straw	36.1	33.8	1.43	0.196
>0.3 mm – 1 mm					
	Concentrate amounts				<0.001
	Fresh grass (pasture)	23.1	23.1	0.691	0.888
	Hay	23.6	22.6	0.695	0.095
	Grass silage	23.3	22.9	0.767	0.495
	Maize silage	23.7	22.5	0.831	0.103
	Straw	23.1	23.1	0.928	0.973
>1mm – 2mm					
	Concentrate amounts				<0.001
	Fresh grass (pasture)	4.15	3.92	0.287	0.373
	Hay	4.03	4.05	0.288	0.908
	Grass silage	4.30	3.78	0.318	0.062
	Maize silage	4.72	3.36	0.345	<0.001
	Straw	3.88	4.20	0.385	0.503
>2mm – 4mm					
	Concentrate amounts				0.076
	Fresh grass (pasture)	4.47	4.72	0.319	0.386
	Hay	4.80	4.39	0.321	0.129
	Grass silage	4.71	4.48	0.354	0.439
	Maize silage	5.64	3.55	0.384	<0.001
	Straw	5.04	4.16	0.429	0.096
>4mm					
	Concentrate amounts				0.117
	Fresh grass (pasture)	2.99	3.51	0.360	0.107
	Hay	3.33	3.18	0.363	0.618
	Grass silage	3.20	3.31	0.400	0.749
	Maize silage	4.22	2.28	0.433	<0.001
	Straw	4.13	2.38	0.484	0.004

[1]The model included the dietary daily amounts of concentrates as a continuous variable, therefore the means for 'yes' or 'no' are not applicable for that variable.

Table 4. Pearson correlations of selected metabolic indicators in milk with proportions of faeces particle fractions (g/kg DM).

Faecal particle	Aceton		Urea		Fat		Fat-protein-ratio	
fractions	coefficient	*P*-value	coefficient	*P*-value	coefficient	*P*-value	coefficient	*P*-value
Sum of all fractions	-0.032	0.576	-0.172	0.002	0.096	0.082	0.010	0.861
>0.3 mm – 1 mm	0.017	0.765	-0.116	0.035	0.074	0.183	0.045	0.416
>1mm – 2mm	0.005	0.936	-0.166	0.003	0.114	0.039	0.022	0.694
>2mm – 4mm	-0.090	0.113	-0.123	0.026	0.024	0.659	-0.073	0.185
>4mm	-0.065	0.247	-0.149	0.007	0.092	0.096	-0.001	0.988

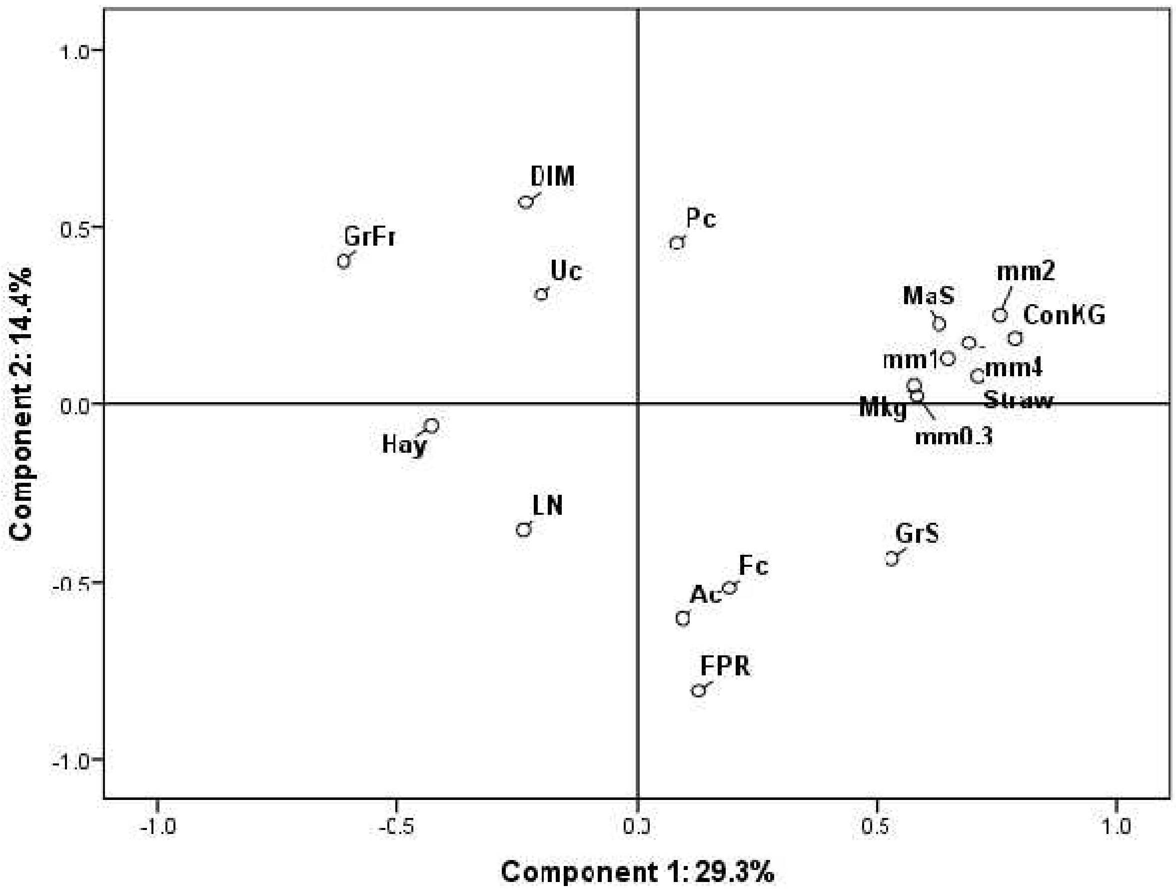

Fig. 1 Plot of the principal component analysis including all diet components and faecal particle fractions as single variables. mm0.3: proportion of particles sized 0.3-1mm; mm1: proportion of particles sized 1mm-2mm; mm2: proportion of particles sized 2mm-4mm; mm4: proportion of particles sized 4mm or larger; ConKG: daily amount of concentrates [kg]; MaS: presence of maize silage; GrS: presence of grass silage; Hay: presence of hay; GrFr: presence of fresh grass; Straw: presence of straw; Mkg: milk yield [kg/day]; Fc: milk fat concentration [g/kg]; Pc: milk protein concentration[g/kg]; Uc: milk urea concentration [mg/100g]; Ac: milk acetone concentration [mg/kg]; FPR: fat-protein ratio in milk; DIM: days in milk; LN: lactation number.

DISCUSSION

Effect of dietary concentrates: The principal component analysis showed that the individual diet components are clearly related to the faecal particle proportions. The correlations show very clearly that increasing use of concentrates was leading to larger DM proportions of particles of all sizes >0.3mm. This is in good agreement with the findings of other research groups (Yang *et al*., 2001; Tafaj *et al*., 2005), indicating that increasing amounts of concentrates may lower the efficiency of fibre digestion in the cow, resulting in more and larger particles in the faeces (Clauss *et al*., 2009). A main factor in this respect is certainly the lower proportion of effective fibre which is a main stimulus for cows to ruminate (Mertens, 1997) and thus to reduce the particle size and increase the surface of the forages. Further, a reduced passage rate might be induced by high dietary fibre, which also improves the digestion rate. This can lead to a physiological trade-off by limiting the intake capacity. However, the particle-sorting ability of the ruminant (Clauss *et al*., 2010) represents a function, which may partly counterbalance this problem.

Effect of maize silage: Maize silage had a similar effect on faecal particles as the concentrates, although the mechanism is probably different: the high influence of maize silage on the proportion of large particles in faeces as found in the current study might be well explained by incompletely digested grains (Lee *et al*., 2002), and only to a lesser degree by the dietary starch (Tafaj *et al*., 2005, 2006) provided by this forage. On the other hand, dietary maize silage might be of value as complementary forage in grass-clover based diets, because it leads to improved nitrogen utilization by the cow and it adds fibre to the diet (Velik *et al*., 2008).

Further factors determining fibre digestion: As far as roughages are concerned, the feeding strategies of the farms in the current study were only classified by indication of the presence of different roughage types like hay or grass silage. A further important factor which should be included in future is a description of the roughage quality. A high quality of roughages which means *inter alia* a moderate fibre concentration, is necessary to maintain high fibre digestion (Tafaj *et al*., 2005; Zebeli *et al*., 2012). Increasing fibre concentrations in forage-only diets lead to reduced intake and digestibility (Leiber *et al*., 2009; Zebeli *et al*., 2012). Finally, also the physical form of roughages, namely the dietary particle size is important for the digestibility (Mertens, 1997; Jalali *et al*., 2012), and this means that not a minimum, but rather an optimum of particle size is required (Rustas *et al*., 2010). Therefore, a practicable roughage quality indication is essentially important to include in on-farm assessments.

Relationships between milk yield and constituents and faecal particle proportions: Milk yield was positively correlated with some of the faecal particle fractions. This is assumed to be rather a confounding effect of the positive relation between milk yields and concentrate amounts, which is the immanent goal and effect of concentrate feeding. In the plot of the principal component analysis, the milk composition was not situated on the same axis as feeding parameters, faecal particles and milk yield, which indicates that neither the chosen approach was able to predict milk composition, nor that the latter is somehow related to fibre digestibility. However, the urea concentration in milk was slightly negatively correlated to all particle fractions, indicating that a low dietary protein supply may also lead to reduced fibre digestion, which is in agreement with the known rumen functions (Van Soest, 1992) and makes clear that sufficient protein supply is necessary for good fibre utilisation.

Conclusions: The current survey has been conducted in order to develop faeces particle evaluation as instrument for on-farm evaluations of feeding situations in roughage-based dairy production systems. The survey revealed that faecal particle proportions responded to the presence of maize silage in the feed and to increasing amounts of dietary concentrates. Thus, the faeces particle sieving might be useful as one component of a novel, digestion based evaluation method for feeding situations in those dairy production systems where low-concentrate diets are the aim. However, indicators of forage quality would be essential for further developments of evaluation systems including faecal particle fractions.

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