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# Effect of Particle Size of Forage in the Dairy Ration on Feed Intake, Production Parameters and Quantification of Manure Index

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ABSTRACT : The objectives of this study were to measure particle size and evaluate the effect of increasing alfalfa hav particle size on production characteristics in lactating Holstein dairy cows. Ninety multiparous Holstein cows in early to mid-lactation were randomly assigned in a complete randomized design for a 30-day period. Animals were offered one of the three diets, which were identical in energy, protein, and chemical composition, but differed only in particle size of alfalfa hay. The treatments were A) total mixed ration (TMR) in which only fine chopped alfalfa hay was incorporated in the ration, B) the same diet in which half of the alfalfa hay was fine chopped and incorporated in the mixed ration and half was long hay and offered as a top dressing, and C) the same diet with long hay alfalfa offered as a top dressing. Distribution of particle size of rations was determined through 20,000, 8,000 and 1,000 µm sieves. The new method of quantitative determination of manure index was examined for each cow on different treatments. The geometric mean length of particle size in the rations was 5,666, 9,900 and 11,549 um for treatments A, B and C, respectively. Fat corrected milk (4%), milk fat percentage and production were significantly different (p<0.05) in treatment A versus B and C (fat corrected milk (FCM, 4%)) 28.3 vs. 35.2 and 32.3 kg/d, fat percentage 2.89, 4.04 and 3.62; but the change of ration particle size had no significant effect on milk production (p>0.05). Blood concentration of cholesterol in treatment A was significantly higher (p<0.05) than treatment B and C (181.0 vs. 150.0 and 155.2 mg/dl). Manure index in treatment C was significantly different (p<0.05) from treatment B (15.86 vs. 17.67). Based on these experimental findings, it is concluded that an increase in the ration particle size can increase milk fat percentage due to providing more physically effective fiber, which in turn could effect changes in manure consistency. (Key Words : Ration Particle Size, Manure Index, Alfalfa Hay, Lactating Dairy Cows)

## INTRODUCTION

Size and shape of ingested forage particles have been reported to affect voluntary intake in sheep (Troelsen and Campbell, 1968) and rate of fermentation and retention time of digesta in growing cattle (Worrell et al., 1986) and dairy cows (Nocek and Kohn, 1988). With respect to the high genetic potential of today's dairy animals, early lactation rations require high levels of energy for peak performance. Forages provide energy as well as fiber to maintain ruminal function and normal milk fat concentration. Physically effective fiber is the fraction of the diet that stimulates chewing and is related to the physical characteristics of neutral detergent fiber (NDF) (primarily particle size) that affect chewing activity (NRC, 2001). To ensure adequate fiber, the NRC (2001) recommends the concentration of total dietary NDF be at least 25% of dietary DM for lactating cows fed diets with alfalfa or corn silage as the predominate forage and dry corn grain. The physical form of specific feeds is usually quantitatively assessed by various sieving methods (Murphy and Zhu, 1997). However, the variety of methods employed has made it difficult to compare results from different laboratories or compile such data into a form that is useful for diet formulation. Particle length of forages can impact on numerous aspects of rumen function and rumen health. A certain amount of long forage particles is important in the diet to ensure proper rumen health by promoting rumination and salivation. Diets with too many small particles may result in feed particles spending less time in the rumen, resulting in less microbial digestion. Therefore, forage particles that are too fine may actually lower feed digestibility and may cause more digestive upsets if they are heavily fed. On the other hand, evaluating the feces or manure can provide information about general health, rumen fermentation, and digestive function of cows (Norgaard, 2006; Norgaard et al., 2007). When feed is not properly fermented in the numen, some

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unfermented nutrients may escape rumen digestion and reach the small intestine. Normally, these nutrients may be digested and absorbed in the intestine, but if the amount is too excessive or the rate of passage is too fast, nutrients may escape both digestion and absorption in the intestine and appear in the feces. Evaluation of manure in the barn is based on a qualitative index of manure and requires more experience by the operator. In addition, it usually is accompanied by error, especially between operators when they evaluate the same herd. The change of qualitative to a quantitative index of manure was one of the aims of the present study.

The first objective of this study was to evaluate the effects of forage particle size on nutrient intake, quality, and quantity of milk production and, secondly, on the change from qualitative manure evaluation to a quantitative index.

# MATERIALS AND METHODS

## **Diets and cow management**

Ninety multiparous Holstein cows, of 607±75 (mean± SD) kg body weight (BW) in the early to mid-stage of the lactation period (104±50 days in milk (DIM)) and 38±8 kg/d milk yield, were used in a completely randomized design to investigate the effects of particle size on quality and quantity of milk production, manure consistency and some blood metabolites. Cows were studied individually during the experimental period. The location of the dairy farm was close to Isfahan, which is located in central Iran (latitude 32°39' North; longitude 51°04' East). The experiment lasted for 30 days with the first 15 days for adaptation and day 15 to 30 used for sampling (urine, blood, and milk). During the experimental period the mean of relative humidity was 28.5% and maximum, minimum and mean temperatures were 25°C, 10.2°C and 18.4°C. respectively. Experimental rations were formulated to meet the requirements according to NRC (2001) for Holstein cows of 610 kg of BW and producing 40 kg of milk per day (Table 1). Rations differed only in the particle size of alfalfa hay. The physical form of forage in ration A was fine chopped forage (alfalfa) mixed with the other part of the ration to form a TMR; in ration B 50% of the forage was fine chopped and mixed with the other part of the ration and the remaining 50% was offered to the cows as a long hay; and in ration C all alfalfa was offered as a long hay. Hay was chopped by TMR wagon blade for 15 min. at four centimeters theoretical length of cut. The long hav alfalfa (whole plant) was top-dressed onto the other part of the ration in the manger before feeding. Experimental diets were fed in equal amounts two times per day. During the experimental period, cows were offered the treatment diets on an ad libitum basis to ensure approximately 10% orts

Table 1	I.	Ingredients	and	nutrient	composition	of	experimental
diets							

		Rations	
-	А	В	С
Dietary components (%)			
Alfalfa hay (long)	-	10.00	20.00
Alfalfa hay (fine chopped)	20.00	10.00	-
Corn silage	19.35	19.35	19.35
Barley grain	21.48	21.48	21.48
Beet pulp	3.87	3.87	3.87
Corn grain	6.90	6.90	6.90
Corn germ	1.15	1.15	1.15
Cotton seed whole	8.06	8.06	8.06
Cotton seed meal	7.38	7.38	7.38
Soy bean	7.38	7.38	7.38
Rumen protected fat	1.73	1.73	1.73
Fish meal	0.77	0.77	0.77
Wheat bran	0.27	0.27	0.27
Limestone	0.38	0.38	0.38
Sodium bicarbonate	0.69	0.69	0.69
Dicalcium phosphate	0.06	0.06	0.06
Salt	0.29	0.29	0.29
Vitamin and mineral mixture	0.24	0.24	0.24
Nutrient composition (% DM)			
Dry matter <sup>2</sup>		68.00	
$TDN^1$		78.00	
ME (Mcal/kg) <sup>I</sup>		2.63	
NEl (Mcal/kg) <sup>l</sup>		1.69	
$CP^2$		17.30	
$RDP^1$		64.00	
Fat <sup>2</sup>		6.60	
$NDF^2$		32.70	
$ADF^2$		24.23	
NFC		36.00	
Cal		0.80	
$\mathbf{P}^{1}$		0.40	

DM = Dry matter; ME = Metabolizable energy; NEI = Net energy for lactation; CP = Crude protein; RDP = Rumen degradible protein as a percentage of CP; NDF = Neutral detergent fibre; ADF = Acid detergent fibre; NFC = Non fiber carbohydrate calculated by difference 100-(% NDF+% CP+% Fat+% ASH); Ca = Calcium; P = Phosphorus.

<sup>1</sup>Calculated according to NRC (2001) for diet component.

<sup>2</sup> Analysed in nutrition laboratory.

(as-fed basis). Animals had access to clean and fresh water at all times, but the amount of water consumption was not measured.

## Particle length in feed, orts and manure

Feed particle size was determined by as-fed sieving. The three sieves which were used for measuring particle size distribution means resulted in three separate rations

	Rations			OT M	Probability
	A B C		- SEM		
Feed (n = 12)					
>20,000 µm	8.76°	33.16 <sup>b</sup>	36.07 <sup>a</sup>	0.505	0.0001
20,000-8,000 µm	41.61 °	45.17 <sup>b</sup>	47.70 <sup> a</sup>	0.790	0.0001
8,000-1,000 μ m	39.45 °	13.38 <sup>b</sup>	12.98 °	0.443	0.0001
<1,000 µm	9.52 *	7.61 <sup>b</sup>	2.60 °	0.127	0.0001
$d_{gw}$	5,630°	9,838 <sup>b</sup>	11,478*	163.9	0.0005
$S_{gw}^{2}$	2.59*	2.49 <sup>a</sup>	2.08 <sup>b</sup>	0.022	0.0087
Particle/gram	576 ª	87 <sup>b</sup>	17°	5.93	0.0001
Surface area <sup>3</sup>	19.78 <sup>a</sup>	12.36 <sup>b</sup>	10.43 °	0.260	0.0001
Ort (n = 12)					
>20,000 µm	31.52 °	34.73 <sup>b</sup>	35.35 °	0.597	0.0001
20,000-8,000 µm	50.71 °	49.46 <sup>b</sup>	45.24 °	0.854	0.0001
8,000-1,000 μm	11.95 <sup>b</sup>	10.71 °	13.99 <sup>a</sup>	0.216	0.0001
<1,000 µm	5.19 ª	4.43 °	4.81 <sup>b</sup>	0.085	0.0001
$d_{gw}$	10,682	11,236	10,669	191.2	0.0494
Sgw	2.25ª	$2.18^{b}$	2.29 ª	0.020	0.0059
Particle/gram	59.0 <sup>b</sup>	40.0 °	68.0 <sup>a</sup>	1.000	0.0001
Surface area	19.61ª	18.12 <sup>b</sup>	19.91 <sup>a</sup>	0.338	0.0481

Table 2. Effect of reducing alfalfa hay particle size on TMR and ort particle size distribution

<sup>1</sup> Geometric mean length as calculated by the ASAE (2003). <sup>2</sup> Standard deviation as calculated by the ASAE (2003).

<sup>3</sup> Surface area of particle size as cm<sup>2</sup>/gram.

SEM = Standard error of the mean. Means with the same letter in each column are not significantly different ( $p \le 0.05$ ).

with four particle sizes (Table 2). Four wooden separator boxes were stacked with a metal screen on top of each other in the following order: sieve with the largest holes (upper sieve = 20,000  $\mu$ m) on top, the medium-sized holes (middle sieve = 8,000  $\mu$ m) next, then the smallest holes (lower sieve = 1,000  $\mu$ m), and the solid pan on the bottom. TMR (500 g as-fed) was placed on the upper sieve and, on a flat surface. sieves were shaken in one direction 5 times then rotated and the separator box turned one-quarter and the process was repeated 7 times, for a total of 8 sets or 40 shakes. After shaking was completed, weight of the material was recorded on each sieve and on the bottom pan.

Feces particle size was determined by dry sieving. The five sieves which were used for measuring particle size distribution meant these sieves separated feces into six sizes (Table 6). These sieves basically were similar to those used for sieving soil. The five metal separators were stacked on top of each other in the following order: Sieve from the largest to smallest holes were 12,500  $\mu$ m, 6,300  $\mu$ m, 3,360  $\mu$ m, 1,190  $\mu$ m, and 850  $\mu$ m, respectively. Around 200 ml of manure was placed in nylon cloth (around 38  $\mu$ m pore size) and washed with warm water and the remaining particles dried in the oven (60°C). Dried manure particles were placed on the upper sieve and the collection of sieves was shaken horizontally for 10 mins. After shaking was completed, the weight of retained material on each sieve

and on the bottom pan was recorded. Data presented in this study includes geometric mean and standard deviation with data assumed to be logarithmically normally distributed, an idea consistent with the ASAE standard (ASAE, 2003). However, the ASAE standard is basically for expressing fineness of feed materials.

## Manure scoring and index

The quantifying index scoring was done for manure consistency evaluation. Manure was excreted as a round shape and the height of the pile was measured by caliper and the diameter of the pile was measured by ruler (at least two measurements). Total volume of the pile was measured in cubic centimeters and the manure index calculated as follow:

Manure index =  $((V/D) \times (V/H))^{0.25}$ 

Where V is volume of pile in  $cm^3$ , D is mean diameter of pile (cm), and H is height of pile in the center (cm).

#### Feed and milk sampling and analysis

Feed offered and orts were measured and recorded daily during the last 10 d of the period to calculate feed intake. Feed samples were collected once weekly, and orts were collected twice weekly for DM determination. Samples were ground through a 1-mm diameter screen. Acid detergent fiber (ADF) analysis was performed according to the method of Van Soest et al. (1991). NDF was determined according to the method of Mertens (2002).

Cows were milked three times daily at 0700, 1500, and 2300 h, with milk weights recorded every day in the milking parlor. Milk samples from each cow were collected at the middle and end of the experiment and preserved with potassium dichromate, stored at 4°C, and sent to the laboratory for milk fat. CP, and lactose determination using a MilkOscan (Funke Gerber, Labortechnik 12105, Berlin-Germany). Because the fat content in milk differed between treatments, milk yield was corrected for 4.0% fat, and energy corrected milk calculated using protein and fat percentage of milk. Blood samples were taken about 3 h after feeding at the middle and end of the experiment from the coccygeal artery or vein into Vacutainers held at 4°C for a maximum of 4 h until serum was harvested and stored at  $-25^{\circ}$ C.

Spot urine samples were also taken at the beginning, middle and end of the experiment by vulval stimulation and analyzed for total nitrogen content by the Kjeldahl method. Urine was analyzed for creatinine using a colorimetric procedure (Darman Kave Kit, Isfahan, Iran). Urine volume was estimated from the creatinine concentration, (Moharrery, 2004).

Urea concentration in urine or serum was measured using the diacetylmonoxime method of Marsh et al. (1957) with a spectrophotometer to measure the change in color at 520 nm wavelength. Serum samples were analyzed for cholesterol by an enzymatic procedure (Kit 10-508, Ziestchem Diagnostic kit, Tehran, Iran).

#### Statistical analysis

The data were analyzed using a general linear model procedure of SAS (2003). The complete randomized model was used to analyze milk yield, milk constituents, urine parameters and blood component data based on the mean values of the samples taken during the experimental period. Feed intake, BW, and DIM were used as a covariant in the model, but because sometime these effects were not significant (p>0.05) these variables were omitted from the model. Duncan's multiple range test (p<0.05) was used to test the significance of difference between means.

## RESULTS

Chemical composition and physical analysis of the total ration is presented in Table 1 and 2. All rations were similar in nutrient composition except for particle size of hay. Ration particle size reflected the amount of chopped alfalfa included in the treatments. Chopping alfalfa resulted in less

**Table 3.** Effect of reducing alfalfa hay particle size on body weight, body weight change, and feed intake in cows on different rations

		Rations		
	А	В	С	SEM
BW (kg)	608	610	607	16.0
BWCH (kg/d)	0.029 <sup>b</sup>	0.108 <sup>a</sup>	0.001 <sup>b</sup>	0.051
DMI (kg)	22.1	23.2	22.2	0.388
N-efficiency	28.4 ª	25.9 <sup>b</sup>	28.1 <sup>a</sup>	0.35

BW = Body weight, BWCH = Body weight change.

BW and BWCH adjusted for body weight in the beginning of experiment. SEM = Standard error of the mean.

Means with the same letter in each column are not significantly different (p<0.05).

material being retained on the 20,000  $\mu$ m screen but increased the amount of particles being retained on both the 1,000  $\mu$ m screen and pan of the separator (p<0.05). Geometric mean increased linearly as the amount of chopped alfalfa in the TMR decreased but standard deviation was increased as geometric mean decreased (p<0.05). The use of chopped alfalfa in ration A significantly increased the number of particles per gram of the ration (p<0.05) and increasing the particles resulted in a higher surface area of each gram of ration (p<0.05). With decreasing percentage of chopped alfalfa in the ration, both the number of particles per gram and surface area of particles were reduced significantly (p<0.05).

The lowest geometric mean particle size for the orts was in the ration with the highest amount of long hay alfalfa (p<0.05), but the standard deviation was increased as geometric mean decreased (p<0.05). The number of particles and the surface area of particles in each gram of orts increased in cows on ration C (p<0.05).

Milk production and composition is summarized in Table 3 and 4. Cows on ration B showed higher body weight change (BWCH) compared to cows on the other treatments (p<0.05). Dry matter intake tended (p = 0.0403) to respond quadratically during the experiment (Table 3). Cows fed ration B (50% long alfalfa hay and 50% fine chopped alfalfa hay) tended to have higher dry matter intake (p>0.05), but the nitrogen efficiency in this group was the lowest compared to the two other groups (p<0.05).

Milk yield and composition in cows on different rations is presented in Table 4. Although milk production increased with increasing particle size, differences were not significant (p>0.05). Fat percentage drastically increased with increasing particle size (p<0.05). The increase in fat percentage tended to respond linearly (p<0.01). Energycorrected milk and fat-corrected milk differed and was highest for cows fed rations containing long hay alfalfa (p<0.05). Milk urea nitrogen (MUN) tended (p = 0.0269) to respond quadratically and cows on ration B showed higher

	Rations			OEM
	Α	В	С	SEM
Milk yield (kg)	37.7	34.9	38.6	2.00
4.0% FCM (kg)	28.3 <sup>b</sup>	35.2 °	32.3 ª	1.37
ECM (kg)	31.84 <sup>b</sup>	36.8 °	34.4 <sup>ab</sup>	1.22
Fat (%)	2.89 <sup>b</sup>	4.04 <sup>a</sup>	3.62 8	0.179
Protein (%)	2.88	2.87	2.80	0.044
Lactose (%)	5.27	5.12	5.32	0.073
TNFS (%)	11.97	12.30	11.98	0.244
MUN (mg/dl)	16.74 <sup>ab</sup>	18.11 <sup>a</sup>	16.35 <sup>b</sup>	0.582

Table 4. Effect of reducing alfalfa hay particle size on milk production and composition in cows on different rations

FCM = Fat corrected milk, ECM = Energy corrected milk, TNFS = Total non-fat solid, MUN = Milk urea nitrogen, SEM = standard error of the mean. Means with the same letter in each column are not significantly different ( $p \le 0.05$ ).

MUN compared to other groups (p<0.05). Other milk components such as protein, lactose and total non-fat solid were not different among rations (p>0.05).

Some parameters in blood serum of cows on different ration are summarized in Table 5. With increasing particle size in the ration, creatinine concentration in blood increased (p<0.05). Cows on ration B (50% long alfalfa hay and 50% fine chopped alfalfa hay) showed higher blood urea nitrogen (BUN) concentration compared to other groups (p<0.05). Cholesterol concentration in the serum of cows on ration A was significantly higher than other groups (p<0.05).

Creatinine and urea-N concentration in urine showed higher values in cows on ration A compared to other groups (p<0.05) and tended to linearly and quadratically respond to ration (Table 5). Total nitrogen excreted via urine (g/d) showed a similar pattern as that seen for MUN (Table 4) and BUN (Table 5).

Total manure physical analysis is presented in Table 6. Ration particle size affected the amount and size of particles in the feces. Both the long alfalfa hay and fine chopped hay increased material retained in 5 out of six screens. There was an increase in the percentage of particles retained on the 841  $\mu$ m screen showing a linear trend (p = 0.0002) as particle size increased in the ration (Table 6). Geometric

mean increased linearly (p = 0.0875) as the amount of chopped alfalfa in the TMR increased but the standard deviation was increased as geometric mean decreased. The use of chopped alfalfa in ration A, significantly decreased the number of particles per gram of feces (p<0.05) and decreasing the particle number resulted in a lower surface area for each gram of ration (p<0.05).

To evaluate manure consistency the manure index has been suggested, which is calculated by a simple measurement of the defecated pile for each cow. Cows on ration B (50% long alfalfa hay and 50% fine chopped alfalfa hay) showed the highest manure index compared to the other groups (p<0.05).

## DISCUSSION

The range of TMR particle size used in the current study is indicative of commercial dairy farms. In this regard, based on a survey composed of over 800 samples, Heinrichs et al. (1999) reported that a minimum of 1% and a maximum of 43% of the particles were observed to be greater that 19.0 mm.

Results of this study indicate that dry matter intake (DMI) during the experiment tended to quadratically respond to the ration. A higher intake was expected for

Table 5. Effect of reducing alfalfa hay particle size on some parameters in blood serum of cows on different rations

	Rations			OEM (
	A	A B		SEM
Blood parameters				
Creatinine (mg/dl)	1.64 <sup>b</sup>	1.79 <sup>a</sup>	1.86 <sup>a</sup>	0.035
N-Urea (mg/dl)	19.26 <sup>ab</sup>	21.50 <sup>a</sup>	18.66 <sup>b</sup>	0.933
Cholestrol (mg/dl)	181.0 <sup> a</sup>	150.0 <sup>b</sup>	155.2 <sup>b</sup>	2.43
Urine parameters				
Creatinine (mg/dl)	114.0 <sup> a</sup>	95.4 <sup>b</sup>	110.0 <sup>a</sup>	3.82
N-Urea (mg/dl)	41.5 <sup>a</sup>	23.0 <sup>b</sup>	40.7 <sup>a</sup>	1.82
Total N (g/d)	218 <sup>b</sup>	237*	213 <sup>b</sup>	5.13

SEM = Standard error of the mean. Means with the same letter in each column are not significantly different ( $p \le 0.05$ ).

	Rations			SEM
	А			
reces				
>12,500 µm	0.12	0.06	0.23	0.073
12,500-6,300 µm	12.18	11.27	12.52	1.375
6,300-3,360 µm	19.58 °	15.91 <sup>b</sup>	18.09 <sup>ab</sup>	1.159
3,360-1,190 μm	43.56	42.03	41.00	1.131
1,190- 841 µm	10.80 <sup>b</sup>	14.44 ª	15.97ª	0.838
< 841 µm	13.68 <sup>ab</sup>	15.80°	11.77 <sup>b</sup>	0.858
gw	2,294	2,009	2,172	93.8
gw 2 gw	2.13	2.17	2.16	0.027
article/gram	3,516 <sup>b</sup>	5,542 °	4,355 <sup>b</sup>	386.0
Surface area <sup>3</sup>	71.22 <sup>b</sup>	81.26 *	75.17 <sup>ab</sup>	2.616
Manure index <sup>4</sup>	17.25 <sup>ab</sup>	17.67*	15.86 <sup>b</sup>	0.545

Table 6. Effect of reducing alfalfa hay particle size on feces particle size distribution in cows on different rations

SEM = Standard error of the mean.

Means with the same letter in each column are not significantly different (p<0.05).

<sup>1</sup> Geometric mean length as calculated by the ASAE (2003). <sup>2</sup> Standard deviation as calculated by the ASAE (2003).

<sup>3</sup> Surface area of particle size as cm<sup>2</sup>/gram. <sup>4</sup> Manure index calculated as: ((Volume of pile/mean pile diameter)×(Volume of pile/height of pile))<sup>0.25</sup>

ration B which contained the same amount of long and fine chopped forage while cows on long hay forage (ration C) and fine chopped forage (ration A) consumed less dry matter. Some studies have reported that reducing forage particle size resulted in increased intake (Rodrique and Allen 1960; Jaster and Murphy 1983; Woodford and Murphy 1986; Fisher et al., 1994; Beauchemin and Rode, 1997), but contrasted with others which reported no differences (Grant et al., 1990; Colenbrander et al., 1991). Beauchemin et al. (1994) reported an interaction (p < 0.01) between forage particle length (alfalfa silage chopped at 5 and 10-mm theoretical length of cut) and percentage of forage in the diet (35 or 65%). In that experiment, DMI was reduced by nearly 3 kg/d when forage percentage was increased from 35 to 65 with diets containing long-chopped alfalfa silage, but was reduced by less than 0.5 kg/d with diets containing short-chopped silage. In the current experiment, cows in early to mid-lactation were used and energy status was improved as feeding diets of long and fine (50:50) chopped forage increased body weight gain. These observations suggest increased energy balance when feeding rations containing both types of long and fine chopped forage, as in ration (B). In agreement with the present study, Teimouri et al. (2004) reported that the amount of BW gain was significantly higher with medium and fine alfalfa treatments compared with long alfalfa treatment. In their study, mid-lactation dairy cows were allotted and energy status was improved as feeding diets of reduced particle size tended to increase BW.

Milk production was not different between treatments (Table 4), but energy-corrected milk and fat-corrected milk showed significant differences between treatments. In this respect, among milk constituents only milk fat percentage showed a significant increase in treatment B ( $p \le 0.05$ ). Milk fat depression was expected in cows consuming fine chopped forage due to low runnial pH and acetic to propionic ratio as observed by Grant et al. (1990) and Fisher et al. (1994). Milk yield was negatively affected by dietary NDF and forage NDF and positively by NFC:NDF ratio, presumably because of their effect on DMI and energy concentration of the diet. Firkins et al. (2001) reported a negative correlation between milk yield and forage NDF. In this regard, Beauchemin et al. (1994) reported that an increase in alfalfa theoretical chop length from 5-mm to 10-mm in low fiber diets increased FCM yield, but in adequate forage fiber diets it decreased FCM yield. Milk protein, lactose, and total non-fat solid (TNFS) were not significantly affected by the dietary factors studied here. However, the milk fat percentage increased linearly with increasing geometric mean particle length of the diet consumed. The result of the present study is in agreement with Leonardi et al. (2005) who reported that particle size distribution had a significant effect on milk composition. In their study, feeding finely chopped oat silage reduced milk fat percentage and yield compared with medium chopped oat silage.

It is generally understood that dairy cattle require fiber in coarse physical form, which is effective in maintaining proper rumen health and function. Current NRC (2001) recommendations state that a minimum mean particle length of 3 mm for alfalfa diets is necessary to maintain rumen pH, chewing activity, and milk fat percentage. However, this recommendation is based on measurements using a vertical sieve shaker containing a profile of wire mesh sieves between 0.30 and 9.5 mm. Because differences between methods of measurements have been reported and

the method of measurement is more applicable to spherical shaped particles, this recommendation is not applicable to measurements made by the Penn State Particle Separator (Murphy and Zhu, 1997). Results of the current study suggest that alfalfa hay-based rations containing about 8.0% of the particles with a size of more than 20,000  $\mu$ m, may be fermented more extensively than those rations which contain more than 30% of the particles with size of more than 20,000 µm, such as rations B and C. The latter rations, maintained normal rumen metabolism and passage rates and resulted to lower milk fat percentage. Although further work is needed to evaluate possible interactions between particle size and ration NDF or NFC level, this study suggests that a level of 9,900 µm for geometric mean particle length of the diet is adequate to maintain rumen health and milk fat percentage for cows in the early to midlactation period.

To my knowledge, this is the first study of the use of different particle size in dairy cows to assess its impact on serum cholesterol (Table 5). The significantly higher concentrations of cholesterol in cows on fine chopped forage may have improved glucogenic metabolites (Kaneene et al., 1997; Duffield et al., 2003) and contributed to improved liver function through reduced liver fat deposition and greater fat export from the liver (higher cholesterol) in these cows.

Manure evaluation includes the assessment of manure appearance and particle size. Diets with a high starch and a low content of physically effective fiber may cause low fiber digestibility, low rumen motility, lack of effective selective retention of long particles in the ruminal system, and fluid manure (Zaaijer and Noordhuizen, 2003). In the present study, different sizes of sieve were used for sieving of the manure. Particle size of 6,300-3,360 µm showed a linear tendency with particle size in the ration. Cows fed fine chopped alfalfa produced feces with a high percentage of particles sized between 6,300-3,360 µm because the rate of passage in this type of ration may be higher than for other rations. In agreement with this result, Bhattia et al. (2008) reported that rate of passage of large orchard grass particles was not influenced by forage source; however, small orchard grass particles had a faster rate of passage when alfalfa was fed with orchard grass compared to orchard grass alone. In this respect, the percentage of particles sized 1.190-841 µm showed an increase (p<0.01) with increasing geometrical means of particles in the TMR. On the other hand, the number of particles per gram of feces increased with increasing particle size in the TMR. Both of these aspects might be the reason for higher motility, chewing and processing action on feed with long particles in the total tract, which resulted in a reduction of particles in the feces. The 95 percentile value (PL95) estimated for feces of cows was 8,630, 8,660, and 1,075 µm for cows on rations A, B, and C, respectively. These values are lower than the PL95 values found from 5 dairy cows in fifteen different Swedish commercial dairy herds (14,000  $\mu$ m: Norgaard et al., 2007) and are higher than the LP95 values in feces from cattle fed forage at maintenance level (6,000  $\mu$ m; Norgaard, 2006). Generally, the large particles and particles containing a high proportion of digestible material are selectively retained in the rumen, whereas the particles containing less digestible material have a higher probability of escaping from the rumen (Huhtanen et al., 2006).

The qualitative index of manure is dependent on the experience of the operator for evaluation and usually is accompanied by error, especially between operators when they evaluate the same herd. The change from qualitative to quantitative index for manure evaluation was one of the aims of the present study (Table 6). The manure index can indicate what type of diet was consumed by cows and to what extent and where it should be digested. Lower values of this index showed a firm pile and a higher index was related to a more runny pile. However, further work is needed to determine possible interactions between particle size and this value to evaluate this index for practical conditions.

# CONCLUSIONS

In the present study increasing geometric mean feed particle size up to 9,900  $\mu$ m of TMR increased DMI in dairy cattle in early to mid-lactation, but the pattern of the relationship was quadratic, meaning that a geometric mean feed particle size of more than 9,900  $\mu$ m in the ration will reduce DMI. With increasing particle size, fat percentage. ECM and FCM drastically improved, but other milk components (such as protein, lactose and TNFS) remained unchanged. The results of the present study showed that particle size in feces have different relationships to particle size in the ration. Under these experimental conditions, particle size in feces longer than 6,300 and 3,360-1,190 was not affected by particle size in the ration.

The new quantitative index for manure evaluation showed a different relationship with particle size in feces. Higher value of this index showed a runny pile and a lower index indicated a firm pile. In this regard, more research is needed for evaluation of this index on commercial dairy farms.

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