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The effects of corn silage particles size and fat supplement on feed intake, digestibility, ruminal function, chewing activity, and performance in mid-lactating Holstein dairy cows

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Abstract An experiment was carried out to determine the effect of corn silage particle size and fat supplements on performance, solid passage rate through the digestive tract, and chewing behavior in dairy cows. The forages were coarse or fine corn silage with geometric means of 8.8±2.7 and 5.6±2.8 mm, and alfalfa with geometrics means of 7.0±3.3 mm, and fat supplements were neutral fat (palm) and soy-oil. Diets were: 1) coarse corn silage and four percent neutral fat supplement, 2) coarse corn silage and four percent soy-oil supplement, 3) fine corn silage and four percent neutral fat supplement, and 4) fine corn silage and four percent soy-oil supplement. The forage to concentrate ratio was 43:57 percent in all diets. A 2×2 factorial balanced change-over design experiment with two replicates was used. Physically effective factor (pef) was affected by corn silage particles size and was higher in coarse corn silage diets, but diets X_{gm} was not affected by silage particles size. Intake of dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), acid detergent fiber (ADF), and non-fibrous carbohydrates (NFC) was not affected by corn silage particles size, fat supplement, and their interactions. However, physically effective NDF (peNDF) intake was significantly higher ($P<0.0107$) in coarse corn silage diets. Digestibility of DM, OM, EE, NDF, and ADF was significantly higher in fine corn silage diets but not affected by fat supplements. Duration of chewing activity was longer in coarse corn silage diets ($P<0.05$). Rumination activities per kg of DM, OM and NDF were longer in coarse corn silage diets. Ruminal solid retention time was longer in coarse corn silage and soy-oil diets ($P<0.05$). Fat yield, total milk solids, and 3.5% fat- corrected milk yield (3.5%FCM) were the highest in the fine corn silage and neutral fat diets. Protein and 3.5% fat-corrected milk efficiencies were higher in diets containing neutral fat supplement. Inert fat supplement can increase energy density of the diets, thus resulting in improved cow performance.

Keywords: fat-corrected milk, non-fibrous carbohydrate, physically effective NDF, soy-oil, total milk solids

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Introduction

The peNDF is an NDF portion of the diet that stimulates chewing activity and establishes the biphasic stratification of ruminal contents (Mertens, 2005). The peNDF is critical for proper ruminal fermentation and animal production. However, interactions of peNDF with dietary ingredients such as protein, supplemented fat and NFC, especially relate to the animal response has not been investigated (Teimouri Yansari et al., 2004; Dewhurst et al., 2006) Researchers indicated that finer particles size of alfalfa hay (Teimouri Yansari et al., 2004; Teimouri Yansari and Pirmohammadi, 2009) and corn silage (Beauchemin et al., 1994; Kononoff and Heinrichs,

2003) caused reductions in both rumination time and milk fat in lactating dairy cows.

Inclusion of fats in the diet during the transition period of early lactation cows has improved the reproductive performance and improved energy balance (Von Soosten et al., 2012), reduced the incidence of metabolic diseases, and allowed energy density to be maintained in diets without increasing the use of rapidly fermentable carbohydrates (Rodney et al., 2015). However, fat supplementation is often accompanied by a decline in dry matter intake (DMI), Fat supplementation in excess of 5% has been implicated in negative effects through its inhibitory effects on ruminal NDF digestibility (Jenkins et al., 1998), acetic and butyric acids concentrations and consequently reduction in milk fat and protein contents in dairy cows (Bauman and lock, 2006; Jenkins et al., 1998). All these modifications generally occur simultaneously, and depend on the amount and the nature of fat and basal diet, the amount of soluble Ca in the rumen, the animal species (Smith et al., 1993), fatty acid profile, supplementation methods, the amount of roughage, and interactions between fat source and feed ingredients (Ueda et al., 2003). The extent of lipolysis of unprotected oils has been estimated to be in the range of 0.85–0.95 (Dewhurst et al., 2006). Reduction in fiber digestibility by dietary fat is due to coating of the fibrous particles that prevents the microorganism attachment, modification in the cellulolytic microbial population, change in microbial cell permeability and reduction in the availability of essential minerals (Ca and Mg) for the microbial activity due to the formation of mineral complexes with the fatty acids (Devendra and Lewis, 1974). Therefore, maximum feeding levels of inert fat sources must be followed to minimize problems with ruminal fermentation.

Associative effects of fat may increase the ruminal mean retention time of NDF and shift the site of its digestion from the rumen to the intestine, possibly due to reducing NDF digestibility (Bauman and Griinari, 2001; NRC, 2001). The understanding of the way in which rumen pool size, rates of ruminal digestion and passage of NDF respond to changes in different levels of peNDF and fat supplements is central to understanding the mechanism by which rumen degradability of NDF changes in the rumen (Lewis et al., 1999).

Fat concentration and/or composition and forage particle size influence milk fat synthesis. The response of lactating dairy cows to rendered fats has not been consistent (Smith et al., 1993), but the reduction in milk and milk fat percentage was higher when rendered animal fats were fed with corn silage based diets compared with alfalfa-based diets (Jenkins et al., 1998). In diets containing corn silage, rumen pH was reduced due to fermentation of the silage starches. As a results, corn silage reduced biohydrogenation or its path changes in the rumen. Trans fatty acids that enter the intestines are absorbed, which can reduce DMI and fat synthesis in the udder (Jenkins et al., 1998; Lock and Bauman, 2007).

The aims of this experiment were to determine the effects of inert fat and soy-bean oil supplements in diets containing coarse and fine corn silage particles size on feed intake, digestibility, chewing activity, gastrointestinal passage rate, and milk yield and milk composition in mid-lactating Holstein cows.

Material and methods

Animals and diets

Eight multiparous lactating Holstein cows averaging 110.0 ± 11.5 days in milk (DIM), and weighing 580.0 ±41.1 kg, were used in this experiment. Cows were cared for according to the guidelines of the Iranian Council of Animal Care (1995). The study was carried out on the Gudosha Dairy Farm, in the city of Bahnamir in Mazandaran province in Iran during fall and winter (2010 and 2011). Four experimental diets were fed in a 2×2 factorial arrangement of treatments in a cyclic change-over design consisting four 22-day periods (adaptation, 14d; sample collection, 7 d; and measurement of chewing activity, 1 d). Forage to concentrate rations was 43:57 percent in all diets and consisting of two levels of corn silage particles size with geometric means of 8.79 ± 2.72 and 5.57 ± 2.82 mm, as coarse and fine particles size, respectively, and two fat supplements (4% of soy-oil or palm fat). Diets were balanced by using Aminocow software, and were: 1) coarse corn silage diet with 4% palm fat, 2) Coarse corn silage diet with 4-% soy-oil, 3) fine corn silage diet with 4 % palm fat, and 4) fine corn silage diet with 4% percent soy-oil supplement (Table 1). Cows were housed in tie stalls and fed *ad libitum* three times daily at 1330, 2230 h and 0630 with TMR, Water was freely available and TMR diets were fed allowing for at least 10% (as-fed basis) refusal.

Corn silage, particle length, and effectiveness of fiber

Corn silage used in the present study was obtained from a local commercial farm. Feed particle size distribution was determined by dry sieving in four replicates using three sets of a Penn State particle separator (Kononoff et al., 2003). The NDF content of the retained material on the sieves of the Penn State particle separator was

Table 1. Ingredients and chemical compositions of four total mixed rations containing two particle size of corn silage and two types of fat supplementation

	Experimental diets			
Particle size	Coarse		Fine	
Fat type	Inert fat	Soy-oil	Inert fat	Soy-oil
Ingredients (%)				
Alfalfa	23.63	23.63	23.63	23.63
Corn silage	22.50	2.50	22.50	22.50
Ground corn	17.84	18.99	17.84	18.99
Ground barley	4.55	4.29	4.55	4.29
Soy-oil meal	9.06	8.69	9.06	8.69
Canola meal	4.93	4.59	4.93	4.59
Cotton seed meal	2.28	2.16	2.28	2.16
Wheat bran	4.93	5.00	4.93	5.00
Fish meal	1.35	1.25	1.35	1.25
Yeast	0.14	0.13	0.14	0.13
Mineral and vitamin premix and buffers[1]	3.35	3.32	3.35	3.32
Molasses	1.44	1.44	1.444	1.44
Inert fat[2]	4.00	0.00	4.00	0.00
Soy-oil	0.00	4.00	0.00	4.00
Chemical composition (%)				
Dry matter	56.47	46.41	56.47	56.41
Crude protein	15.79	15.71	15.79	15.71
Total fat	7.59	7.94	7.52	7.94
Neutral detergent fiber	32.08	31.97	32.08	31.97
Acid detergent fiber	17.78	17.78	17.78	17.78
NFC[3]	35.31	36.27	35.31	36.27
Calcium	0.98	0.98	0.98	0.98
Phosphorus	0.60	0.60	0.60	0.60
NE (Mcal/kg)[4]	1.62	1.66	1.62	1.66

[1] Contained 40% NaCl, 25% Dynamite, 10% K, 7% Mg, 12%S, 1,000 mg Fe/kg, 2% $ZnSO_4.H_2O$, 2% $MnSO_4.4H_2O$, 0.01% $CoSO_4.6H_2O$, 0.009% Na_2SeO_3, 0.012% ethylenediamine dihydroiodide, 0.8% $CuSO_4.5H_2O$, 680,000 IU/kg of Vitamin A, 160,000 IU/kg of Vitamin D, and 2,000 IU/kg of Vitamin E.

[2] RP10 inert fat powder (Malaysia) that consisted of 85 and 2% palmetic and stearic acid, respectively.

[3] NFC (% of dry matter) =100-(Ash (%)- CP (%) –EE(%) – NDF(%)) (NRC, 2001).

[4] Net energy for lactation, calculated using the Amino cow® Software based on NRC (2001).

measured (Van Soest et al, 1991). Dietary pef of the TMRs was determined based on the proportion of DM retained on three 19-, 8-, and 1.18-mm sieves (Kononoff et al., 2003). The peNDF was calculated by multiplying the NDF content of each portion on each sieve by the pef. The geometric mean (X_{gm}) of particle size (mm) and the standard deviation of the geometric mean (SD_{gm}) were calculated (Table 2) according to ASAE S424.1 (2002).

Body Weight, intake, and apparent total-tract nutrient digestibility

Body Weight was measured weekly and at the end of each period. The DMI of the cows was measured daily. Daily samples of TMRs, residuals, and total feces were collected daily for 7 d of each period at 00630, 01330 and 2230 h, weighed, and 7 days samples were composited and subsampled. The samples were dried at 55°C for 48 h and ground using a Wiley mill (1-mm screen), analyzed for DM, Kjeldahl N, ether extract, OM, ash at 605°C for 3 h (AOAC, 2002), ADF, and NDF (Van Soest et al., 1991; with α-amylase, heat-stable, Product Number A 3306 Sigma-Aldrich added for concentrates during NDF extraction; without sodium sulfite). The NDF was expressed without residual ash. Non-fibrous carbohydrates were calculated by using the following equation: [100- (% CP + % NDF + % ash + % EE)] (NRC, 2001). Using the chemical components of TMRs and feces, the feed intake, and digestibility of nutrients were calculated. Intake of peNDF (kg/d and % of DMI) was calculated for each treatment (Kononoff et al., 2003).

Rumen fermentation and kinetics

The digestion kinetics were measured using Cr-mordanted

Table 2. Particle size distributions (material retain on each sieve,% of DM), physically, and geometric mean of four total mixed rations containing two particle size of corn silage and two types of fat supplementation

	Experimental diets				SEM	P-values		
Particle size	Coarse		Fine			Particle size	Fat type	Interaction
Fat type	Inert fat	Soy-oil	Inert fat	Soy-oil				
Material retain on each sieve,% of DM								
19.0 mm	21.71^{a}	22.69^{a}	14.76^{b}	11.29^{c}	1.11	<0.01	0.22	0.05
8.0 mm	17.79	16.51	22.11	23.36	3.42	<0.05	0.99	0.56
1.18 mm	48.40^{a}	48.69^{a}	39.83^{b}	42.72^{b}	1.21	<0.01	0.25	0.30
pan	10.93^{b}	11.67^{b}	23.53^{a}	20.33^{a}	3.03	<0.01	0.50	0.30
Pef[1]	0.88^{a}	0.88^{a}	0.79^{b}	0.77^{b}	0.01	<0.01	0.20	0.19
PeNDF[2]	28.40^{a}	28.47^{a}	24.52^{b}	25.18^{b}	0.43	0.01	0.69	0.31
X_{gm}[3] (mm)	2.82	2.98	2.57	2.71	0.29	0.35	0.63	0.90
SD_{gm}[4] (mm)	0.32	0.41	0.54	0.60	…	…	…	…

[a, b, c] Means within a row with common subscript do not differ ($P > 0.05$).

[1] pef: physically effective factor, determined as the proportion of DM retained on three sieves of Penn State particle separator (Kononoff, 2002).

[2] The peNDF was calculated by multiplying NDF content of each portion on each sieve on each pef.

[3] Geometric mean particle that was calculated using ASAE (S424.1, 2002).

[4] Standard deviation of the geometric means of particles, using ASAE (S424.1, 2002).

SEM: Standard error of the mean.

NDF of wheat straw as the single-dose marker for solid passage rate measurement. The Cr-mordanted fiber was prepared by mordanting ground wheat straw through a 5-mm screen using a Wiley mill (Uden et al., 1980). Each cow was orally fed with a mixture of 2 kg of concentrates and 250 g of the marker before morning feeding on the 15th d of each period. Fecal grab samples were collected at 0, 6, 10, 12, 14, 18, 22, 26, 30, 36, 42, 48, 54, 60, 72, 84, 96, 120, and 144 h after dosing from the rectum to determine the passage rate, ruminal, and total mean retention time, and time delay (transit time) of the marker. Samples were dry-ashed, and fecal Cr concentrations was determined by direct current plasma emission spectroscopy (AOAC, 2002). Fecal Cr excretion curves were fitted to the double compartment model, as represented by the following two exponential constants and a time delay (Grovum and William, 1973):

$$Y = Ae^{-k_1 (t-TT)} - Ae^{-k_2 (t-TT)},\ k_1 = k_2 \text{ for } t \geq T$$
$$Y = 0 \text{ for } t = TT$$

in which Y = marker concentration (ppm); A = scale parameter; k_1 = ruminal passage rate (%/h); k_2 = lower digestive tract passage rate (%/h); t = sampling time post dosing (h); and TT = transit time or time delay of marker. The total mean retention time was calculated as the sum of ruminal mean retention time ($1/k_1$), and for the lower digestive tract, mean retention time ($1/k_2$) plus the transit time. Parameters were estimated by NLIN regression using the PROC NLIN (iterative Marquardt method) of the SAS® (2005). The estimated parameters were analyzed according to a previously described experimental design.

Eating, ruminating and chewing times

Eating and ruminating activities were monitored visually over a 24-h period at d 22 of each treatment period. Eating and ruminating activities were recorded at 5-min intervals and each activity was assumed to persist for the entire interval. A period of rumination was defined as at least 5 min of ruminating activity, followed by at least 5 min without ruminating activity. Total time spent on chewing was calculated as the total time spent eating and ruminating (Teimouri Yansari et al., 2004).

Milk production

Milk production was recorded daily. On d 18 to 23 of each period, milk samples were collected at each morning and evening milking. Approximately 100 mL of each milk sample were composited and analyzed for total solid, total protein, fat, and lactose (AOAC, 2005). The 3.5% FCM (Britt et al., 2003) and 3.5% FPCM, (Leiva et al., 2000) were calculated as follows:

3.5% FCM = (16.23 × milk fat kg) + (0.432 × milk kg)
3.5%FPMC= (12.82 × milk fat, kg) + (7.13 × milk protein, kg) + (0.323 × milk, kg)

Statistical analysis

The data on particles size were analyzed as a completely randomized design for the effects of the diets using the REML variance component and PROC MIXED of SAS® (2005). Mean separation was performed using the PDIFF procedure and significance was declared at $P<0.05$. The geometric mean and its standard deviation were calculated using ASAE S424.1 (ASAE, 2002). Using PROC MIXED of SAS® (2005), the experimental data were analyzed as a 2 × 2 factorial experiment in a change-over design using the following model:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + S_k + Cow_{k(j)} + \gamma_m + \alpha\times\beta_{j\,i} + e_{ijklm}$$

in which Y_{ijkl} is the dependent variable; μ is overall mean; α_i is fixed effect of particle size (i =1 and 2); β_j is fixed effect of supplementals fat (j =1 and 2); $S_{k,}$ is the effect of squares (k =1 and 2); $Cow_{k(j)}$ is the random effect of cow within square; γ_m is the effect of periods; $\alpha\times\beta_{j\,i}$ is the fixed effect of interaction between particle size and supplemental fat and e_{ijklm} is the residual error. Means were separated using the Tukey's multiple range tests at P<0.05.

Results and discussions

Particle size distributions and physical effectiveness of diets

The particle size of corn silage significantly changed the pef, and peNDF content of the TMR diets (Table 2). Dry matter on three sieves, pef, and peNDF were significantly higher in coarse diets than fine diets. The X_{gm} was similar in all diets (Table 2) but dry matter remaining on the pan was higher in fine corn silage diets (P<0.01). In addition, fat supplementation and interaction of particle size and fat types had no significant effect on particle size distribution, and consequently on the pef, and peNDF content of the diets. Kononoff et al. (2003) and Zebeli et al. (2010) suggested the use of Penn State Particle Separator (PSPS) to express the particle size distribution and to calculate the content of peNDF in the diets of dairy cows. Kononoff and Heinrichs (2003) and Yang and Beauchemin (2004) found that peNDF measured using the PSPS was a poor predictor of chewing time and (or) ruminal pH for cows fed corn silage- and alfalfa silage-based diets. However, Mertens (1997) and Zebeli et al. (2006) proposed that dietary peNDF should be maintained at or above 21% and 23% to ensure a ruminal pH greater than 6.0 and 6.1, respectively. In this experiment, the mean peNDF

Table 3. Daily intake of dry and organic matter, nutrients, their digestibility, and ruminal kinetic of four total mixed rations containing two particles size of corn silage and two types of fat supplementation

	Experimental diets				SEM	*P*-values		
Particle size	Coarse		Fine			Particle size	Fat type	Interaction
Fat type	Inert fat	Soy-oil	Inert fat	Soy-oil				
Daily intake (kg)								
Dry matter	21.86	23.53	20.42	20.52	2.12	0.08	0.44	0.49
Organic matter	19.43	21.56	18.72	18.95	1.90	0.09	0.37	0.50
Crude protein	3.45	3.70	3.22	3.22	0.33	0.80	0.48	0.49
Ether extract	1.66	1.70	1.65	1.63	0.17	0.09	0.14	0.46
Neutral detergent fiber	6.99	7.55	6.53	6.58	0.68	0.06	0.17	0.64
peNDF[1]	5.94^{ab}	6.93^{a}	4.92^{b}	5.13^{b}	0.42	<0.01	0.77	0.49
Acid detergent fiber	5.25	5.76	4.78	4.69	0.69	0.07	0.56	0.41
Non Fiber carbohydrate[2]	5.95	5.79	5.44	5.47	1.13	0.31	0.87	0.79
Digestibility (%)								
Dry matter	65.15^{c}	66.04^{bc}	70.23^{ba}	72.24^{a}	4.26	<0.01	0.49	0.84
Organic matter	66.95^{c}	67.91^{bc}	71.79^{ba}	72.24^{a}	4.16	<0.01	0.36	0.78
Crude protein	68.59	69.71	73.78	71.88	7.14	0.07	0.70	0.59
Ether extract	61.27^{ab}	57.06^{b}	71.98^{ab}	74.02^{a}	13.60	<0.01	0.69	0.49
Neutral detergent fiber	48.39^{b}	57.26^{ab}	60.28^{a}	58.72^{a}	9.06	<0.05	0.37	0.08
Acid detergent fiber	39.94^{b}	44.65^{ab}	51.35^{a}	51.41^{b}	8.12	<0.01	0.39	0.25
Non fiber carbohydrate	93.44	93.52	93.32	95.73	7.14	0.95	0.40	0.41
Ruminal kinetics								
Ruminal passage rate (%/h)	4.65	4.35	4.90	4.45	0.39	0.06	0.74	0.07
Lower compartment passage *rate (%/h)*	3.15	3.23	3.48	3.33	0.28	0.06	0.48	0.09
Ruminal mean retention time (h)	21.50^{ab}	22.91^{a}	20.41^{b}	22.47^{a}	0.75	<0.05	<0.05	0.08
Hind gut retention time (h)	31.77	31.71	28.79	38.26	8.00	0.59	0.43	0.42
Time delay (h)	25.25	24.50	26.00	27.00	1.32	0.33	0.41	0.35
Total mean retention time (h)	78.50	78.45	75.14	79.50	2.45	0.53	0.51	0.42

$^{a, b, c}$ Means within a row with common subscript do not differ (*P*> 0.05).

[1] peNDF: physical effective NDF calculated as ration NDF multiplied by cumulative proportion of particles retained on 3 sieves of Penn State Particle Separator (Kononoff et al., 2003).

[2] NFC (% of dry matter) =100-(Ash (%)- CP (%) –EE(%) – NDF(%)) (NRC, 2001). SEM: Standard error of the mean.

values of coarse and fine silages were around 28.4 and 24.8, respectively, and indication of optimal conditions in the rumen.

Intakes and nutrient digestibility

Corn silage particle size, type of fat supplements and interaction of silage particles size and fat supplements had no significant effect on intake of DM and other nutrients, except for peNDF intake ($P < 0.01$; Table 3). Onetti et al. (2003) reported that reducing the particle size of corn silage from 32 to 19 mm increased the dry matter and NDF intakes and positively affected the animal performance. The response of cow to decreases in particle size depends on the proportion of concentrates in diets and interactions between nutrients. Zebeli et al. (2006) reported that reducing dietary hay particle size from 28.7 to 9.2 mm increased DMI in sheep by 13% only at a low-concentrate level (13% in DM), but when a high-concentrate diet (~40% in DM) was fed no differences were observed in sheep. In this experiment, diets contained 54% concentrate (Table 1), therefore, reducing of forage particle size had no effect on the intake of DM and other nutrients. Intake of peNDF was higher in coarse corn silage diets ($P<0.01$) that is due to the higher peNDF content of coarse corn silage diets in comparison to other diets. There was a significant negative correlation between peNDF and NDF intake in experiments in which reduction in particle size significantly increased the dry matter intake (Teimouri Yansari et al., 2004).

As a general principle, dietary supplemental fat reduces the feed intake in ruminants (NRC, 2001, Onetti and Grummer, 2004). Onetti and Grummer (2004) reported that 3.1 to 3.8 percent supplemented animal fat and calcium salt of fatty acid decreased DMI about 0.90 to 0.97 kg per day in cows. Grant and Weinder (1992) found that while the addition of 11.6% whole raw soy-oils in alfalfa silage decreased daily DMI by approximately 7.1%, but the reduction in DMI was greater for fine silage than for coarse silage diets. Jenkins et al. (1998) found that consumption of 5% saturated animal fat had no significant effect on DMI in fine and coarse alfalfa silage diets because inert fat has limited effect on biohydrogenation, and the rumen fermentation (Garnsworthy, 2002). In this experiment, the NDF content of diets was about 32% in DM, which may have prevented the negative effect of fat supplements on the intake (Grant and Weinder, 1992).

Digestibility of DM and OM (except crude protein) was significantly higher in fine corn silage diets ($P<0.01$, Table 3). Crude protein digestibility was not affected by silage particles size, type of supplemental fat and their interaction. Digestibility of NDF, ADF, and EE was higher in fine corn silage diets ($P<0.05$). Lewis et al. (1999) and Pantoja et al. (1994) found reducing forage particles size increased the surface area for microbial attachment, ruminal microbial digestion, and fermentation, and increased volatile fatty acid production. Beauchemin et al. (1994) and Yang and Beauchemin (2006) found that reducing dietary forage particle size increased ruminal degradability of protein and OM and decreased escapable protein because the reduction of the particle size increases the surface area for microbial attachment and digestion (Miron et al., 2001).

Fat supplements did not impact on the nutrients digestibility although fiber digestion and ruminal fermentation are often negatively associated with the feeding of dietary fat. Feeding fat supplementations reduced DMI and ruminal NDF digestibility, and reduction increases as unsaturated fat level increases (Lewis et al., 1999). Ruminal digestibility of DM and NDF of alfalfa hay was reduced by over 20% when soy-oil was added to the diet (Palmquist and Jenkins 1980). However, the undesirable effect of fat may be compensated at the lower compartment of the digestive tract. However, Lewis et al. (1999) reported that tallow fed at 5% of dietary DM disrupted ruminal fermentation, but had no negative effects on total tract nutrient digestion. The negative effects of particles size on digestibility of nutrients in corn silage diets with soy-oil and inert fat supplements were the result of the coating of the fibrous particles that prevents attachment of microorganisms to the food particles (Devendra and Lewis, 1974).

Rumen particulate passage rate and retention time

Ruminal mean retention time was affected by fat supplement types and silage particles size (Table 3) and was higher in soy-oil and coarse silage diets ($P<0.05$). But Ruminal particulate passage rates, lower compartment passage rate, hindgut retention time, time delay and total mean retention time were not affected by silage particles size and fat supplement types. We expected that ruminal particulates passage rate would increase in fine silage particles size, but it seems that coating of fine particles with fat and oil was decreased ruminal digestion and declined particles size reduction process in the rumen, and ultimately increased ruminal mean retention time as equal as coarse particles size diets. The ruminal digesta outflow is the principal factor that limits feed consumption in ruminants. To improve forage intake, the rate of removal from the rumen must be increased by increasing the rate of digestion, the rate of passage, or both (Bhatti et al., 2008).

Tafaj et al. (2007) found that mean ruminal passage rates was 5 to 7 %/h (average 6 %/h) when cows fed diets with large particles. In one experiment, mean solid rumen passage rate was 4.06 percent per hour (Khalile Argomandi and Teimouri Yansari, 2011) and slower than our experiment because in this experiment, the main forage in the diets was alfalfa and its rumen passage rate is slower than corn silage; and unlike corn silage alfalfa hay needs water absorption (Teimouri Yansari et al., 2004).

Chewing activity

Eating time (minutes per day) tended to be longer for coarse silage diets (P<0.06), due to higher peNDF level (Table 2). According to Kononoff and Heinrichs (2003), eating time was not affected by corn silage particle sizes, because there were no differences in DMI between fine and coarse corn silage diets.

Corn silage particles size and fat types had no significant effect on rumination time (minutes per day). Generally, NDF intake was increased by reducing peNDF in diets. There is a positive relationship between NDF intake and chewing activity (Teimouri Yansari et al., 2004), but in this experiment, DM intake was not affected by silage particle size and fat types. Intake of peNDF was affected by silage particle sizes (P<0.01) and was higher in coarse corn silage diets (P<0.05) due to the higher peNDF level in coarse silage diets (Table 2).

Rumination time per kg DM, OM and NDF was longer for fine corn silage diets (Table 5), It is likely that on these diets, rumen pH was reduced and rumination was higher to compensate the reduction in pH (Aschenbach et al., 2010). Under normal conditions, 80% of diet particles that are crushed and broken are attributed to chewing and the rest to microbial activity. If secretion of saliva is reduced during eating fine corn silage (eating time per kg of DM, OM and NDF were not affected by corn silage particle size), it would be compensated by more rumination to increase the ruminal pH (Zebeli et al., 2006). Rumination per kg NDF was affected by the fat type and was higher in soy-oil supplemented diets because ruminal mean retention time was higher for inert fat supplemented diets (Table 3).

Chewing activity (minutes per day) was longer for coarse corn silage diets (Table 4). Chewing is the sum of eating and rumination activities, and eating activity tended to be higher for coarse corn silage diets (P<0.06). Chewing activity per kg of NDF tended to be higher for fine corn silage diets (P<0.07), due to greater rumination time per kg of NDF in these diets (P<0.01). According to Kononoff and Heinrichs (2003), chewing activity per kg of NDF was longer for the diets containing coarse corn silage, because of more NDF was consumed on coarse corn silage diets.

Milk production, composition, and efficiency

Whole milk production was not affected by corn silage particle size and type of fat supplements (Table 5).

Table 4. Chewing activity of cows fed on four total mixed rations containing two particles size of corn silage and two types of fat supplementation

	Experimental diets				SEM	P-values		
Particle size	Coarse		Fine			Particle size	Fat type	Interaction
Fat type	Inert fat	Soy-oil	Inert fat	Soy-oil				
Chewing activity (min/d)								
Eating	380.6^{ab}	435.6^{a}	353.4^{b}	358.8^{ab}	70.00	0.06	0.18	0.42
Rumination	473.1	503.1	437/9	450.0	43.16	0.75	0.20	0.15
Total chewing activity	853.7^{ab}	926.9^{a}	817.9^{b}	808.6^{b}	70.16	<0.05	0.06	0.11
Eating activity per kg of								
Dry matter	18.23	18.63	19.78	20.16	4.96	0.58	0.88	0.99
Organic matter	19.99	20.34	19.80	21.73	4.76	0.80	0.66	0.76
Neutral detergent fiber	57.04	58.08	56.69	62.58	7.071	0.56	0.45	0.72
Rumination activity per kg of								
Dry matter	22.33^{b}	23.90^{ab}	27.25^{ab}	27.51^{a}	2.56	<0.05	0.38	0.86
Organic matter	22.33^{b}	23.90^{ab}	24.92^{ab}	24.51^{a}	2.43	0.05	0.27	0.92
Neutral detergent fiber	60.47^{b}	63.79^{ab}	67.80^{ab}	72.09^{a}	11.58	<0.01	<0.05	0.11
Chewing activity per kg of								
Dry matter	38.60	39.45	40.97	45.83	5.76	0.18	0.36	0.51
Organic matter	42.31	43.07	44.77	49.24	6.04	0.41	0.28	0.37
Neutral detergent fiber	126.8	122.9	128.2	135.1	15.49	0.07	0.17	0.46

$^{a, b, c}$ Means within a row with common subscript do not differ (P> 0.05). SEM: Standard error of the mean.

Table 5. Milk production, compositions, and efficiency of cows that fed four total mixed rations containing two particles size of corn silage and two types of fat supplementation

	Experimental rations				SEM	P-values		
Particle size	Coarse		Fine			Particle size	Fat type	Interaction
Fat type	Inert fat	Soy-oil	Inert fat	Soy-oil				
Milk production(kg/day)								
Milk	30.77	30.53	28.77	30.25	2.48	0.05	0.74	0.55
3.5-FCM[1]	30.51^{ab}	29.73^{ab}	30.91^{a}	26.82^{b}	3.36	0.82	<0.05	0.08
3.5-FPCM[2]	30.59	29.37	30.99	26.91	4.37	0.35	0.16	0.55
Milk composition and yield								
Fat (%)	3.42^{ab}	3.23^{ab}	3.94^{a}	2.84^{b}	0.69	0.40	0.25	0.10
Fat yield (kg)	1.06^{ab}	1.00^{ab}	1.14^{a}	0.86^{b}	0.22	0.99	<0.05	0.18
Protein (%)	3.01	3.17	3.01	3.17	0.21	0.67	0.14	0.50
Protein (kg)	1.00^{a}	0.97^{a}	0.86^{b}	0.96^{a}	0.09	<0.05	0.51	0.10
Lactose (%)	4.56	4.53	4.65	4.74	0.27	0.18	0.94	0.71
Total solid (%)	12.83^{b}	12.58^{b}	14.01^{a}	12.18^{b}	1.05	0.16	0.01	0.06
Solid not fat (%)	8.64^{a}	8.55^{ab}	8.27^{b}	8.65^{b}	0.31	0.60	0.50	<0.05
Milk efficiency								
Milk	1.43	1.34	1.40	1.42	0.13	0.80	0.34	0.33
3.5-FCM	1.43	1.30	1.48	1.29	0.19	0.49	0.03	0.57
3.5-FPCM	1.41^{ab}	1.29^{ab}	1.53^{a}	1.26^{b}	0.22	0.40	<0.05	0.32

$^{a, b, c}$ Means within a row with common subscript do not differ (*P*> 0.05).

[1] 3.5% FCM= (16.23 × milk fat kg) + (0.432 × milk kg) (Britt et al., 2003).

[2] 3.5%FPMC= (12.82 × milk fat, kg) + (7.13 × milk protein, kg) + (0.323 × milk, kg) (Leiva et al., 2000).

SEM: Standard error of the mean.

However, 3.5% FCM yield was higher due to inert fat supplement diets (P<0.01), because of fat production (% and kg) was significantly higher on these diets. Chalupa and Ferguson (1986) reported milk yield increase of 3 to 8 percent (1 to 2.5 kg) per 0.45 kg added fats. Fats and fatty acids have negatively affect the ruminal digestion, especially fiber digestion and enhancement of rumen passage rate, resulting in a reduction in feed efficiency, as a result of toxic effects on the cellulolytic bacterial community caused by the medium chain and unsaturated fatty acids (Onetti et al., 2001). The inert fat supplement containing 85% palmitic acid had no negative effect on rumen fermentation, and passed through the rumen, absorbed in the intestine and transferred to milk with high efficiency. Firkins and Eastridge (1994) and Warntjes et al. (2008) found that milk production was increased by adding palm fat supplement while milk fat decreased. Jenkins (2011) reported that unsaturated fatty acids could have a dual effect on ruminal biohydrogenation in that they modify the microbial population and increase the amount of substrate that must be biohydrogenated. Supplementation of the diets with soy-oil that is high in linoleic acid increased the risk of milk fat depression via the rumen unsaturated fatty acid load (Jenkins, 2011). Milk fat (yield and percentage) was higher in fine corn silage diets that supplemented with the inert fat. Supplements that are high in saturated fat (palmitic and stearic) do not increase the risk of milk fat depression; however, supplementation of calcium salts of fatty acids can reduce milk fat (Harvatine and Allen, 2006; Lundy et al., 2004). In the current experiment, milk fat yield and percent were higher in inert fat supplemented diets than soy oil supplement diets. Pantoja et al. (1994) showed that FCM was decreased by increasing unsaturated fat supplements (milk fat decreased), due to decreased fiber digestion and acetate and butyrate production (Garnsworthy, 2002).

Milk fat yield and percentage were lower on fine corn silage diets supplemented with soy-oil than the other diets. Protein yield was lower on fine corn silage diets containing inert fat (P<0.05). Milk lactose percentage was not affected by silage particle size and type of fat supplement similar to other reports (Onetti et al., 2001, 2003; Teimouri Yansari et al., 2004; Zebeli et al., 2006). Total milk solid content was higher on diets supplemented with inert fat (P<0.05), due to higher milk fat yield and percentageon these diets. Milk efficiency of 3.5 FCM and 3.5% and fat- and protein-corrected milk (3.5% FPCM) were greater for inert fat than the soy-oil supplemented diets (P<0.5). Reduction in milk fat and protein may be due to decreased salivary secretion on fine corn silage diets (Sharifi et al., 2012), decreased ruminal pH, modification of biohydrogenation pathways and production of intermediate fatty acids (trans 10 cis 12 18:2) that have inhibitory effect on fatty acid synthesis in the mammary gland (Lock and

Bauman, 2007; Lock et al., 2007; Overton et al., 2007). Supplemental fats can greatly disrupt fermentation in the rumen, causing reduced digestibility of non-lipid energy sources. Ruminal digestion of structural carbohydrates can be reduced, and this reduction in digestion is accompanied by reduced production of methane, hydrogen, and VFA, including lower acetate to propionate ratio (Jenkins, 2011; Lock and Bauman, 2007). With supplemental fat, the proportion of energy supply from dietary starch, propionate and glucose production decreased (Cabrita et al., 2007; Garnsworthy, 2002), gluconeogenesis from amino acid increased, and amino acid availability for milk protein synthesis decreased (Garnsworthy, 2002; Grant and Weinder, 1992; Jenkins and Jenny, 1992). However, Bouattour et al., (2008) and Whitlock et al. (2003) found that milk solid was increased by adding soy-oil to dairy goats and cattle diets. In this experiment, DMI was not affected by the type of fat supplement; therefore, the increased efficiency was due to increases in 3.5% FCM on inert fat supplemented diets ($P<0.05$). Grant and Weinder (1992) found that DMI was decreased by adding 11.6% raw soy-oil to fine and coarse alfalfa silage diets, but 4% FCM production was not different between the two diets due to increased milk fat production. However, 4% FCM efficiency was higher on fine alfalfa silage diets containing fat supplement. Jenkins et al. (1998) examined the effect of two levels of animal fat (0 and 4 % of DM) and particles size of alfalfa silage and found that milk production efficiency increased by adding four percent fat supplement.

Conclusions

Digestibility of DM, OM, NDF and ADF was higher on fine corn silage than on coarse silage diets. The 3.5%-FCM and 3.5%-FPCM production were higher in palm fat supplemented cows. This showed that the palm fat had no negative effects on ruminal microbial function. Therefore, palm fat by increasing the NEL content of the diet, could be a good supplement to enhance the corrected milk fat production and efficiency. Negative effects of soy-oil were greater on 3.5%-FCM and milk efficiency in diet containing fine corn silage. This indicates the importance of high peNDF diets when unsaturated fats are added to the dairy diets.

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چکیده آزمایشی به منظور تعیین اثر اندازه ذرات ذرت سیلویی و مکمل‌های چربی بر گوارش پذیری، نرخ عبور از دستگاه گوارش، رفتار مصرف خوراک و عملکرد گاوهای شیرده، انجام شد. میانگین هندسی اندازه ذرات ذرت سیلویی درشت و ریز و یونجه خشک به ترتیب ۸/۸±۲/۷، ۵/۶±۲/۸ و ۷/۰۱±۳/۳ میلی‌متر بود و مکمل‌های چربی پالم و روغن سویا بودند. جیره‌ها عبارت بودنداز :۱) جیره ذرت سیلویی درشت و چهار درصد مکمل چربی پالم، ۲) جیره ذرت سیلویی درشت و چهار درصد روغن سویا ۳) جیره ذرت سیلویی ریز و چهار درصد مکمل چربی پالم و ۴) جیره ذرت سیلویی ریز و چهار درصد روغن سویا. در تمام جیره‌ها نسبت علوفه به کنسانتره ۴۳ به ۵۷ درصد بود. داده‌ها در قالب طرح چرخشی متعادل و چینش فاکتوریل با دو تکرار برای تجزیه داده‌های آزمایش واکاوی شدند. عامل موثر فیزیکی تحت تاثیر اندازه ذرات ذرت سیلویی قرار گرفت و در جیره‌های ذرت سیلویی درشت بیشتر بود، اما میانگین هندسی جیره‌ها تحت تاثیر اندازه ذرات ذرت سیلویی قرار نگرفت. مصرف ماده خشک، ماده آلی، چربی خام، الیاف نامحلول در شوینده‌ی خنثی و اسیدی و کربوهیدرات‌های غیر الیافی تحت تاثیر اندازه ذرات ذرت سیلویی و نوع مکمل چربی قرار نگرفتند. اما مصرف الیاف موثر فیزیکی در جیره‌های دارای ذرت سیلویی درشت بیشتر بود. گوارش پذیری ماده خشک و آلی، چربی خام، الیاف نامحلول در شوینده‌ی خنثی و اسیدی در جیره‌های داری ذرت سیلویی ریز بیشتر بود، اما تحت تاثیر نوع مکمل چربی قرار نگرفت. فعالیت جویدن در جیره دارای ذرت سیلویی درشت بیشتر بود. فعالیت نشخوار به ازای هر کیلو ماده خشک، ماده آلی و الیاف نامحلول در شوینده خنثی در جیره‌های ذرت سیلویی درشت بیشتر بود. نرخ توقف مواد جامد در شکمبه در جیره‌های ذرت سیلویی درشت بیشتر بود. تولید چربی، کل مواد جامد شیر و شیر تصحیح شده برای چربی در جیره ذرت سیلویی ریز و مکمل چربی پالم از دیگر جیره‌ها بیشتر بود. بازده شیر تصحیح شده برای ۳/۵ درصد چربی و برای پروتئین در جیره‌های دارای مکمل چربی پالم بیشتر بود. مکمل چربی پالم غلظت انرژی جیره‌ها را افزایش داد، لذا عملکرد گاوهای شیرده بهبود یافت.