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Effect of different dietary ratios of corn silage to barley silage on fiber digestibility, feeding behavior, and milk yield performance of dairy cows

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corn silage (CS) on nutrient utilization, feeding behavior, and performance of dairy cows. Eight Holstein lactating cows were used in a replicated 4 × 4 Latin square design, and the cows were offered diets containing different ratios of CS to BS (22% of DM) as 1) 3:0 (CS₃BS₀); 2) 2:1 (CS₂BS₁); 3) 1:2 (CS₁BS₂); and 4) 0:3 (CS₀BS₃). The diets were iso-energetic and iso-nitrogenous, but the dietary concentration of indigestible neutral detergent fiber (iNDF) and proportion of long particles (>19 mm) were increased with increasing BS proportion in the diets. The degradability characteristics of the silages were evaluated by in situ technique. Both silages were similar in predicted total tract NDF digestibility (TTNDFD = 35 %), 30-h undegradable NDF (uNDF30h = 32 %), NDF digestion rate (kd = 1.8 %/h), but CS was lower in iNDF and higher non fibrous carbohydrates (NFC). Intake of DM was unchanged, but NDF intake was linearly increased with increasing BS levels. Total tract NDF digestibility did not differ among treatments, but DM digestibility linearly decreased with increasing BS proportion. Feeding different ratios of CS: BS did not affect milk yield or milk fat and protein concentration. Furthermore, ruminating time was unaffected by dietary treatments but increasing BS level decreased quadratically the eating and total chewing times. The experimental treatments did not affect plasma β -hydroxybutyric acid and glucose concentrations, but BS increased linearly ruminal ammonia and blood urea nitrogen concentration. The results indicated that BS had a higher iNDF content and a negative impact on DM digestibility and N utilization. However, partial or total substitution of CS with BS led to similar DM intake and milk yield, possibly as a result of similar fiber digestion rate and

Abstract This study assessed the substitution effects of barley silage (BS) for

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Introduction

As a result of increased milk production of dairy cows during recent years, cows are fed low forage and high concentrate diets to meet their high nutritional require-

uNDF30h.

ments (Migliorati et al., 2017; Kahyani et al., 2019). However, a minimum level of dietary effective fiber from forage sources is necessary to maintain health and performance of dairy cows (NRC, 2001). Dietary fiber from

forage source remains longer in the rumen and induces mastication, ensalivation, ruminal motility, mat formation, and filling (NRC, 2001; Mertens, 2016), thereby affecting the intake, feeding behavior, fiber digestibility, milk fat percentage, and overall animal health (Zebeli et al., 2012). In many areas, alfalfa and corn silage (CS) are commonly fed to high producing dairy cows. The high fiber digestibility of these forages could reduce the physical gut fill, allowing the animal to consume more feed and produce more milk (Oba and Allen 1999a). These crops grow best at high summer temperatures and require frequent irrigation in spring and summer. However, increasing demands for water and the variable drought conditions make the prospects of water scarcity a rapidly growing concern in many regions (Molden, 2013). Compared to the conventional feeding system, the autumn-winter cereals such as barley need lower irrigation water, and due to higher rainfall in autumn and winter, barley is less susceptible to drought. Such production systems allow the optimization of water resources (Tudisco et al., 2010).

Several studies have investigated the nutritional value and fiber digestibility of different varieties of barley silage (BS) as well as the effect of maturity on performance of dairy cows (Wallsten and Martinsson, 2009; Nair et al., 2016; Refat et al., 2017a). In general, BS contains higher amounts of protein, NDF, and lignin, but lower levels of ether extract and starch compared to CS (Benchaar et al., 2014; Refat et al., 2017a). Most previous research indicated that the substitution of BS for CS, on dry matter (DM) basis (Benchaar et al., 2014; Refat et al., 2017a; Hosseini et al., 2019) or grass silage (Ahvenjarvi et al., 2006) reduced the dry matter (DM) intake and milk yield. The decreases in milk yield were attributed to decreases in nutrient digestibility and supply to the animal. In contrast, Migliorati et al. (2017) showed that milk yield and quality did not differ between diets containing BS and CS. These authors, however, did not report the nutrient intake and digestibility. The main reason for this discrepancy may be related to the original forage NDF content and digestibility. These parameters in the silages are widely affected by the species, variety, maturity, growing environment, and the mechanical and ensiling processing (Ferraretto and Shaver, 2012; Refat et al., 2017a). The importance of NDF digestibility of the silages has been verified on DM and energy intake and animal performance (Dado and Allen, 1996; Nair et al., 2016). The greater NDF digestibility results in faster disappearance from the rumen, reduced ruminal fill, and greater voluntary feed intake especially in high-producing dairy cows (Oba and Allen, 1999a). The NDF digestibility depends mainly on the content of indigestible NDF (iNDF) and potentially digestible NDF (pdNDF) and its fractional rate of digestion and passage (Huhtanen et al. 2007). The undegraded NDF at 288 h of in situ incubation (uNDF_{288h}; Huhtanen et al., 2007) are used as the laboratory measure of iNDF. The iNDF or uNDF_{288h} will not be nutritionally available to microbial digestion and set the final extent of rumen fiber digestion (pdNDF, Mertens, 2016). Nevertheless, the uNDF measured at 288 h of in situ incubations did not biologically reflect retention time and ruminal fill in dairy cows. Oba and Allen (1999b) indicated that high producing dairy cows have a fiber retention time of 30 to 36 h. Therefore, the forage uNDF value after 30-h incubation (uNDF_{30h}), as the single time point incubation, may be a decent estimate of the total mean retention time (West et al., 1997), gut fill limits of uNDF (Oba and Allen, 1999b), and pdNDF digestion rate (Lopes et al., 2015). It is also used for evaluating fiber digestibility and forage quality (Oba and Allen, 1999a; Refat et al., 2017a).

Results from previous studies revealed that NDF digestibility of CS and BS could affect the performance of dairy cows (Benchaar et al., 2014; Refat et al., 2017a; Hosseini et al., 2019), and the in vitro total tract NDF digestibility (TTNDFD) model could accurately be used to predicte the rate of fiber digestion and NDF digestibility in vivo (Lopes et al., 2015). Other studies (Kahyani et al., 2019) showed that adjustment of diets differing in forage sources for NDF digestibility (uNDF_{30h} content) resulted in similar DM intake and milk yield in dairy cows. This study aimed to compare the substitution effects of BS for CS on nutrient digestibility and intake, feeding behavior, and milk yield in lactating cows. We hypothesized that partial or complete substitution of BS for CS in the diets adjusted for NDF digestibility (predicted total tract NDF digestibility or uNDF_{30h}) would not negatively affect digestibility and DMI, and thereby milk yield in dairy cows.

Materials and methods

In situ NDF degradation

The experiment was conducted from February to May 2017, at the dairy facilities of the Lavark Research Station (Isfahan University of Technology, Isfahan, Iran). Wholecrop barley was cultivated in the university farm with a semi-arid climate and significant variations between winter and summer temperatures (32° 32′ N, 51° 23′ E, 1630 m above sea level). The corn plants were obtained

Table 1. Chemical composition, physical and fiber degradation parameters of corn and barley silages

Item¹ Corn silane Barley silage

_ltem¹	Corn silage	Barley silage	SEM
Chemical composition,			
% of DM (unless otherwise stated)			
DM (% of as fed)	27.6 ^b	36.9 ^a	0.56
OM (% of DM)	92.9 ^a	88.8 ^b	0.69
CP (% of DM)	7.82 ^b	10.7 ^a	0.39
EE (% of DM)	2.82	2.52	0.039
NFC ² (% of DM)	29.0	21.4	-
NDF (% of DM)	53.2	54.2	0.91
ADF (% of DM)	30.4	30.8	0.20
ADL (% of DM)	5.37	5.60	0.28
NE _L ³ (Mcal/kg of DM)	1.41	1.25	-
Ammonia-N (% of DM)	0.33	0.50	-
рН	3.90	4.30	-
Particle size distribution, mm pore size			
19.0 mm (% of DM)	14.0 ^b	49.0 ^a	1.96
8.0 mm (% of DM)	60.0a	28.9 ^b	2.32
1.18 mm (% of DM)	23.8a	19.8 ^b	0.107
<1.18 (% of DM)	2.20	2.36	0.689
Geometric mean of particle size (mm)	9.98 ^b	14.0 ^a	0.250
Fiber characteristics			
Physically effective NDF _{8mm} (% of DM)	39.4 ^b	42.2 ^a	0.30
Physically effective NDF _{1.18mm} (% of DM)	52.0	52.9	0.57
NDFd _{30h} ⁴ (% of NDF)	42.0	43.6	1.22
NDFd _{30h} (% of DM)	22.3	23.6	1.11
uNDF _{30h} ⁵ (% of NDF)	59.4	58.2	1.77
uNDF30h (% of DM)	32.4	31.6	0.45
iNDF ⁶ (% of NDF)	19.5 ^b	23.8ª	0.29
iNDF (% of DM)	10.4 ^b	12.9 ^a	0.54
pdNDF ⁷ (% of NDF)	80.5ª	76.2 ^b	0.54
pdNDF (% of DM)	42.8 ^a	41.3 ^b	0.29
pdNDF degradation rate (%/h)	1.79	1.82	0.03
Effective degradability (% of NDF)	39.9	40.1	0.27
TTNDFD ⁸ (% of NDF)	35.9	34.3	0.55
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¹DM: Dry matter; OM: Organic matter; CP: Crude protein; EE: Ether extract; NFC: Non fibrous carbohydrate; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin; NE_L: Net energy for lactation

from a commercial farm. Both plants were harvested at the dough stage of maturity and ensiled in bunker silos for at least 3 months before being used in the experiment. The chemical composition and particle size distribution of the silages are presented in Table 1. Two ruminally-cannulated non-lactating Holstein cows were fed a total mixed ration (TMR) containing 23 % chopped alfalfa hay, 24 % CS, 23 % wheat straw and 25 % concen-

trate twice daily at 8:00 and 18:00 h. The in situ technique was used to determine the NDF degradation characteristics of CS and BS. Samples of CS and BS, as well as TMR, were dried in a forced-air oven at 60 °C for 48 h and ground to pass a 1 mm screen by a Wiley mill before in situ incubation. All samples (0.4-0.5 g) were weighed into polyester bags (5×4 cm; with a pore size of $25 \mu m$), sealed, and incubated in triplicate for 288, 240, 120, 96,

 $^{{}^{2}}NFC = 100 - [CP + NDF + fat + ash].$

³Based on NRC (2001).

⁴NDFd_{30h}: NDF degradation after 30-h incubation.

⁵uNDF_{30h}: NDF residue after 30-h in situ incubation.

⁶iNDF: NDF residue after 288-h in situ incubation.

⁷pdNDF: potentially degradable NDF determined by 288-h in situ incubation

⁸TTNDFD: predicted total-tract NDF digestibility using in situ TTNDFD model (Lopes et al., 2015).

 $^{^{}a,b}$ Means within a row with different superscripts differ (P < 0.05).

72, 48, 36, 30, 24, 12, and 6 h. After removal, the bags were rinsed for 14 min in tap water in a washing machine for total clearing, and then transferred to forced ventilation oven (60 °C), where they were kept for 48 h. Indigestible NDF and uNDF_{30h} concentrations were determined by in situ incubation of samples for 288 h and 30 h, respectively. Total-tract NDF digestibility (TTNDFD, + kp)] $\{0.9, \text{ as described by Lopes } et al. (2015). The$ amount of pdNDF was calculated from the difference between NDF and iNDF. The kd of pdNDF was calculated from NDF residue measurements taken at 0, 6, 12, 24, 30, 36, and 48 h of *in situ* incubation in the ruminal fluid. The kp of pdNDF was predicted 2.67 %/h and the hindgut digestion of NDF was assumed to be 10 % of total NDF digestion (Lopes et al., 2015). The effective degradability of each feed was calculated as: ED = A + B[kd/(kd + kp)], where A is the soluble fraction.

Cows, management, and treatments

Eight multiparous Holstein cows ($82 \pm 10 \text{ d in milk}$; mean \pm SD) were assigned to treatment sequence in a replicated 4×4 Latin square design. Treatment periods were 21 d, with the last 7 d for data collection. At the begin-

ning of the experiment, body weight (BW) of cows was 639 ± 76 kg with a body condition score (BCS) of 3.3 ± 0.45 and milk production of 58.9 ± 4.5 kg/d. Animals were transferred to individual pens 2 weeks before starting the experiment and received the farm diet for adaptation to the pen. Cows were blocked into 2 squares of 4 cows each based upon the parity (=2 and >2). Treatments were randomly assigned after blocking to minimize carryover effects. The diet was supplied twice daily at 09:30 and 17:30 h, allowing for 5-10 % refusals, and with free access to fresh water.

Cows were housed in individual pens $(4 \times 4 \text{ m})$ in an open-side barn. Cows were offered diets containing different ratios of CS to BS (22% of DM) as 1) 3:0 (CS₃BS₀); 2) 2:1 (CS₂BS₁); 3) 1:2 (CS₁BS₂); and 4) 0:3 (CS₀BS₃) as represented in Table 2. The diets were formulated according to the Cornell Net Carbohydrate and Protein System, version 5.0 for similar net energy for lactation (NE_L), crude protein (CP) concentration and to meet the requirements of a multiparous cow producing 55 kg milk/d with 28 g/kg milk protein and 30 g/kg fat, assuming a DM intake of 28 kg/d. All diets contained 36:64 forage to concentrate ratio.

Cows were milked 3 times daily (09:00, 17:00 and

Table 2. Ingredients of the experimental diets (% of DM)

ltem	Corn silage: Barley silage							
item	3:0	2:1	1:2	0:3				
Corn silage	22.0	14.8	7.20	0.00				
Barley silage	0.00	7.20	14.4	22.0				
Alfalfa hay	14.0	14.0	14.0	14.0				
Beet pulp	3.80	3.80	3.80	3.80				
Barley grain, ground	12.0	12.0	12.0	12.0				
Corn grain, ground	24.0	24.4	25.2	25.0				
Soybean meal	6.80	6.40	6.00	5.80				
Extruded soybean	7.92	7.92	7.92	7.92				
Canola meal	5.20	5.20	5.20	5.20				
Rice bran	1.00	1.00	1.00	1.00				
Fat powder ¹	0.40	0.40	0.40	0.40				
Sodium bicarbonate	0.80	0.80	0.80	0.80				
Calcium carbonate	0.80	0.80	0.80	0.80				
Magnesium oxide	0.20	0.20	0.20	0.20				
Vitamin-mineral premix ²	0.40	0.40	0.40	0.40				
Mineral premix ³	0.40	0.40	0.40	0.40				
Salt	0.30	0.30	0.30	0.30				

¹ Fatty acids including C12:0 (2 g/100 g fatty acids), C14:0 (5 g/100 g fatty acids), C16:0 (80 g/100 g fatty acids), C18:0 (2 g/100 g fatty acids), C18:1 (7 g/100 g fatty acids), and C18:2 (3 g/100 g fatty acids).

²DM basis: 1,300,000 IU/kg vitamin A; 360,000 IU/kg vitamin D3; 12,000 IU/kg vitamin E; 10 g/kg manganese; 16 g/kg zinc; 4 g/kg copper; 0.15 g/kg iodine; 0.12 g/kg cobalt; 0.8 g/kg iron; and 0.08 mg/kg selenium.

³DM basis: 0.1 g/kg cobalt; 4.5 g/kg cu; 13.5 g/kg manganese; 18 g/kg zinc; 0.2 g/kg iodine; 0.072 g/kg selenium; 55 g/kg magnesium; 245 g/kg calcium.

01:00 h), and milk yield was recorded during the last 5 d of each period. Milk was sampled every other day and samples were preserved with potassium dichromate, and held at 4 °C pending analysis. Milk samples were analyzed for fat, protein, lactose, and total solids (Milk-O-Scan 134 A/B Foss Electric HillerØd, Denmark). The yield of 3.5 % fat corrected milk (FCM, kg/d) was calculated as: 3.5 % FCM (kg/d) = 0.432 (kg milk) + 16.23 (kg fat). Feed efficiency was calculated by dividing the daily actual or corrected milk production (kg/d) by DM intake (kg/d). Body condition score (BCS) was scored by two experienced scorers at the beginning and the end of each experimental period. Also, at the end of each experimental period, back-fat thickness (BFT) was measured at an imaginary line between the hooks and pins at the sacral examination site, once weekly by ultrasound (SonoVet 600V; BCF Technology Ltd., West Lothian, UK). Cows were weighed on 2 consecutive d immediately after the morning milking at the beginning and the end of the trial; daily BW changes were measured before the morning feeding.

Sampling and analyses

During sampling in each period, the amount of feed offered and refused was recorded to calculate DM intake. Samples of the TMR were collected at feeding and orts collected before morning feeding. The samples were refrigerated until the end of the collection period, at which time they were combined by period (and by cow for the orts), sub-sampled and then stored at -20 °C for later analysis. The samples were dried at 100 °C in a forcedair oven for 24 h (AOAC International, 2002; method 925.40) and ground to pass a 1-mm screen using a rotary mill (Wiley mill, Arthur H. Thomas). Duplicate subsamples were analyzed for CP (AOAC method 955.04, 2002), ether extract (AOAC method 920.39, 2002), and NDF (using heat stable alpha-amylase), acid detergent fiber (ADF), and acid detergent lignin (ADL) (Van Soest et al., 1991). Organic matter was determined by ashing at 550 °C overnight. Additional samples of forages, TMR, and individual orts of each cow were taken for particle size separation during the same time of each experimental period. All samples were frozen immediately at -20 °C until analyses. After thawing, particle size distributions of the representative sub-samples (in duplicate) were determined on as fed basis using the Penn State Particle Separator (PSPS; NASCO, Fort Atkinson, WI) equipped with 3 sieves (19, 8, and 1.18 mm). After separation, the DM of each separated fraction was determined by oven drying at 60 °C for 48 h. The physical effectiveness factor (pef) was determined as the DM proportion of particles retained on 2 sieves (pef>8) and on 3 sieves (pef>1.18) of the PSPS, respectively. The physically effective NDF of 2 (peNDF>8) and 3 (peNDF>1.18) sieves were calculated by multiplying the NDF concentration of the feed by the fraction on pef>8 and pef>1.18, respectively (Beauchemin and Young, 2005). Actual intakes of peNDF, with both systeCS, were then calculated after adjustment for this value in orts. The geometric mean particle length (GMPL) was calculated according to the ASAE (1995; method S424.1).

Fecal samples were collected approximately every 9 h from d 16 to 19 so that 8 samples were taken from each cow each period. The samples were composited by cow and kept at -20 °C until analysis. These samples were dried in a forced-air oven at 60 °C for 72 h, ground to pass through a 1-mm screen, and analyzed for starch, DM, ash, NDF, and CP, as described above. Apparent total-tract digestibilities of nutrients were determined using acid-insoluble ash as an internal marker (Van Keulen and Young, 1977).

Ruminal and blood parameters

Blood samples were collected, in tubes containing EDTA (2.1 mg/mL), from the coccygeal vein 4 h after morning feeding on d 19 in each period. Blood plasma was separated after centrifugion at 3000 × g at 4 °C for 15 min, and frozen at -20 °C until analysis. Blood urea nitrogen (BUN) and glucose concentrations were measured using commercial kits (Pars Azmoon Co., Tehran, Iran) according to the manufacturer's instructions. Plasma concentrations of β -hydroxybutyric acid (BHBA) were determined by an enzymatic method (Randox Lab. Ltd, UK). On the last d of each period, ruminal fluid samples were taken 4 h after morning feed delivery, using a stomach tube connected to a vacuum pump. The initial 50 mL of aspirated ruminal fluid was discarded to minimize contamination with saliva. The pH of ruminal fluid was immediately measured by using a portable, digital pH meter (SN: 137243, Hanna Instruments, Portugal). Samples were then strained through two layers of cheese-cloths. The ruminal fluid sample for NH₃-N analysis was centrifuged at 30,000 × g for 20 min at 4 °C; the supernatant was removed and analyzed for ammonia by the colorimetric phenol-hypochlorite method (Broderick and Kang, 1980).

Feeding behavior

Eating and ruminating behaviors were monitored visu-

ally for a 24-h period on d 20 of each period. Chewing activity was noted every 5 min, and each activity (i.e., eating, ruminating, resting) was assumed to persist for the entire 5-min. Total chewing time was determined as the sum of total eating and ruminating times. The sorting index was calculated as the ratio of actual intake to expected intake of particles retained on each sieve of the pen state particle separator (PSPS, Leonardi and Armentano, 2003). The predicted intake of an individual fraction was calculated as the product of the DM intake of the total diet multiplied by the DM percentage of that fraction in the fed TMR. A sorting index of 100, >100, and <100 indicated no sorting, sorting for, and sorting against, respectively.

Statistical Analyses

Data were analyzed as a duplicated 4×4 Latin square design using PROC MIXED of SAS (2002, SAS Institute Inc., Cary, NC). The model included the fixed effects of square, period within square, and treatment. Cow within square was included in the RANDOM statement. The statistical model used for analyses was:

$$Y_{ijkm} = \mu + S_m + P(S)_{im} + A(S)_{jm} + T_k + e_{ijkm}$$

where, Yijkm = each observation, μ = overall mean, S_m = fixed effect of square m, $P(S)_{im}$ = fixed effect of period i within square m, $A(S)_{jm}$ = random effect of cow j within square m, T_k = fixed effect of treatment k, and e_{ijkm} = random error. The differences among the treatment means were evaluated using the Tukey-Kramer test. Polynomial orthogonal contrasts were used to evaluate the linear, quadratic, and cubic effects of the level of BS in the diets. Significance was declared at $P \le 0.05$, and trends were noted if $0.05 < P \le 0.10$.

Results

Fiber digestibility and chemical characteristics

The chemical composition and physical characteristics of BS and CS are presented in Table 1. The CS had lower DM (276 vs. 369 g/kg as-fed), CP (81 vs. 107 g/kg DM), iNDF (104 vs. 129 g/kg DM), peNDF_{>8} (394 vs. 422 g/kg DM), NH₃-N (0.33 vs. 0.50 g/kg) and pH (3.9 vs. 4.3) than BS. In contrast, organic matter (OM, 929 vs. 888 g/kg DM), EE (28 vs. 25 g/kg DM), NFC (290 vs. 214 g/kg DM), and pdNDF (805 vs. 762 g/kg NDF) were higher in CS compared to BS. There was no difference in NDF (537 g/kg DM), ADF (306 g/kg DM), ADL (55 g/kg DM), NDFd_{30h} (428 g/kg NDF), uNDF_{30h} (320 g/kg DM), NDF digestion rate (0.018 h⁻¹), and TTNDFD (351 g/kg NDF)

between CS and BS.

As BS contained less calculated NE_L (1.25 vs. 1.41 Mcal/kg) and more CP (107 vs. 78 g/kg DM) compared with CS, dietary contents of corn grain and soybean meal were slightly adjusted (Table 2) to balance the dietary NE_L and CP concentrations across treatments (Table 3). Water was added to the diets to adjust the dietary DM content because of higher DM content in BS than CS. The 30-h NDFd, uNDF_{30h}, NDF digestion rate, and TTNDFD were not different among the treatments (Table 3). However, the iNDF and pdNDF₂₈₈ (P=0.08) as a percentage of DM tended to increase and decrease linearly, respectively, with increasing the BS level in the diets.

Physical characteristics of the silages and TMR offered are shown in Tables 1 and 3, respectively. The particles retained on 19 mm sieve (140 vs. 485 g/kg DM) and geometric mean particle size (Xgm, 9.98 vs. 14.0 mm) were lower in CS than BS, but the proportion of particles retained on 8 mm (289 vs 600 g/kg DM) sieve was greater for CS compared with BS. Consequently, the particles retained on 19 and 8 mm sieves were increased and decreased, respectively, as BS replaced CS in the diets. The Xgm, peNDF_{>1.18}, and peNDF_{>8} were not affected by the dietary treatments.

Nutrient intake and digestibility and ruminal pH

Nutrient intake and total tract digestibility are presented in Table 4. The DM and NEL intakes were not different among the treatments, but intakes of NDF (P<0.01), iNDF (P<0.01), and uNDF_{30h} (P=0.02) increased in a linear and cubic fashion with increasing BS level in the diets. Total-tract digestibility of DM (P=0.01) and OM (P=0.02) decreased linearly as BS replaced for CS. No differences were observed in NDF total tract digestibility across the dietary treatments. No differences were observed for ruminal pH values across the treatments, whereas the level of ruminal ammonia-N was elevated (P=0.01) with increasing inclusion of BS in the diet.

Milk yield and composition and blood metabolites

Average milk yield (54.2 kg/d) and FCM (47.6 kg/d) did not differ among the treatments (Table 5). Moreover, the treatments had no effect on milk composition including fat (2.76 %), protein (2.86 %) and lactose (4.64 %). Feed efficiency was constant among treatments, and cows produced 1.76 and 1.53 kg milk or FCM per kg of DM intake. The BW, BCS, and back fat thickness were similar in cows fed different ratios of BS to CS.

Table 3. Chemical composition, physical characteristic, and fiber digestibility of the experimental diets

Table 3. Chemical composition, physical characteristics	Corn silage: Barley silage						
Item ¹	3:0	2:1	1:2	0:3	SEM		
Chemical composition							
DM (% of as fed)	58.8	58.4	58.1	57.5	-		
CP (% of DM)	15.9	15.8	15.8	15.9	-		
EE (% of DM)	4.74	4.27	4.57	4.15	-		
NFC ² (% of DM)	38.5	39.6	38.1	37.7	-		
NDF (% of DM)	31.8	32.1	32.5	32.8	-		
Forage NDF (% of DM)	19.0	19.1	18.9	19.2	-		
ADF (% of DM)	16.1	16.5	16.8	16.3	-		
ADL (% of DM)	3.31	3.61	3.89	3.16	-		
Metabolizable protein g/day	2815	2799	2785	2773	-		
NE _L ³ (Mcal/kg of DM)	1.60	1.59	1.59	1.59	-		
Particle size distribution							
19.0 mm (% of DM)	6.15 ^c	8.57 ^b	11.3 ^a	12.6ª	1.58		
8.0 mm (% of DM)	18.8ª	15.6 ^b	13.4 ^c	13.5 ^c	0.44		
1.18 mm (% of DM)	44.3	42.7	42.9	45.8	4.13		
<1.18 (% of DM)	30.7	33.2	32.4	28.1	4.57		
Geometric mean of particle size (mm)	3.33	3.24	3.38	3.72	0.29		
Fiber characteristics							
Physically effective NDF _{8mm} (% of DM)	7.51	7.92	8.00	8.33	0.46		
Physically effective NDF _{1.18mm} (% of DM)	20.8	21.9	22.1	22.6	0.97		
uNDF _{30h} ⁴ (% of NDF)	50.1	51.2	51.8	51.4	0.89		
uNDF _{30h} (% of DM)	17.8	17.9	18.6	18.1	0.75		
iNDF ⁵ (% of NDF)	24.8 ^c	25.3 ^{bc}	26.7 ^{ab}	27.7 ^a	0.38		
iNDF (% of DM)	7.76 ^d	8.28 ^c	8.68 ^b	9.04ª	0.08		
pdNDF ⁶ (% of NDF)	75.2ª	74.7 ^{ab}	73.3 ^{bc}	72.3 ^c	0.38		
pdNDF (% of DM)	24.0	23.8	23.8	23.8	0.08		
NDF degradation rate (%/h)	3.87	3.92	3.92	4.45	0.43		
Effective degradability (% of NDF)	45.4	44.9	43.7	44.8	0.68		
TTNDFD ⁷ (% of NDF)	48.4	48.3	47.6	47.2	0.57		

¹DM: Dry matter; CP: Crude protein; EE: Ether extract; NFC: Non fibrous carbohydrate; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin; NE_L: Net energy for lactation. ²NFC = 100 − [CP + NDF + fat + ash]. ³Based on NRC (2001). ⁴uNDF_{30h}: NDF residue after 30-h in situ incubation. ⁵iNDF: NDF residue after 288-h in situ incubation. ⁶pdNDF: potentially degradable NDF determined by 288-h in situ incubation. ⁷TTNDFD: predicted total-tract NDF digestibility using in situ TTNDFD model (Lopes et al., 2015). ^{a,b,c,d}Means within a row with different superscripts differ (P < 0.05).

Table 4. Nutrient intake, total tract digestibility, and ruminal pH in lactating cows fed different ratios of corn silage to barley silage

Item ¹	Corn silage: Barley silage				SEM	<i>P</i> -value ²		
item	3:0	2:1	1:2	0:3	SEIVI	L	Q	C
Intake								
DM (kg/d)	30.9	31.6	31.2	31.5	0.78	0.53	0.65	0.21
NE∟ (Mcal/d)	49.2	50.4	49.7	50.2	1.31	0.55	0.63	0.21
NDF (kg/d)	9.49	10.2	10.1	10.4	0.25	< 0.01	0.20	0.02
uNDF _{30h} (kg/d)	5.51	5.65	5.81	5.71	0.14	0.02	0.11	0.79
iNDF (kg/d)	2.40	2.61	2.71	2.85	0.07	< 0.01	0.30	0.02
Apparent total tract digestibility								
DM (%)	72.6	68.0	69.0	66.8	1.34	0.01	0.36	0.02
OM (%)	74.1	69.7	70.6	68.8	1.32	0.02	0.41	0.02
NDF (%)	48.0	42.2	44.7	43.5	2.67	0.38	0.34	0.16
Ruminal								
рН	6.26	6.47	6.30	6.52	0.172	0.43	0.98	0.14
ammonia N (mg/dL)	12.7	11.7	14.0	15.6	1.14	0.01	0.22	0.72

 1 DM: Dry matter; NE_L: Net energy for lactation; NDF: Neutral detergent fiber; uNDF $_{30h}$: NDF residue after 30-h in situ incubation; iNDF: NDF residue after 288-h in situ incubation; OM: organic matter.

²L: linear effect, Q: quadratic effect, C: cubic effect.

Table 5. Milk production and composition, feed efficiency, body measurements, and blood me-

tabolites in lactating cows fed different ratios of corn silage to barley silage

Item ¹	Corn silage: Barley silage				_		<i>P</i> -value ²	2
ILCIII	3:0	2:1	1:2	0:3	SEM	L	Q	C
Yield								
Milk (kg/d)	53.4	55.0	53.8	54.7	1.88	0.68	0.77	0.23
FCM (kg/d)	46.5	48.4	47.9	47.5	1.26	0.49	0.16	0.35
Milk composition								
Fat (%)	2.71	2.78	2.85	2.72	0.137	0.76	0.18	0.65
Protein (%)	2.86	2.90	2.87	2.86	0.026	0.43	0.18	0.36
Lactose (%)	4.61	4.68	4.63	4.62	0.052	0.69	0.18	0.31
Feed efficiency ³								
Milk/DMI	1.74	1.76	1.73	1.74	0.068	0.79	0.76	0.71
FCM/DMI	1.51	1.54	1.54	1.51	0.033	0.98	0.16	0.98
Body measurement changes								
BW (kg/d)	0.33	0.94	0.57	0.27	0.272	0.62	0.127	0.55
BCS (/21 d)	-0.08	-0.15	-0.12	-0.04	0.099	0.75	0.425	0.95
BFT (mm/21 d)	-0.75	-1.50	-1.25	-0.38	0.990	0.75	0.425	0.95
Blood metabolites								
Glucose (mg/dL)	65.1	64.1	61.1	66.8	3.97	0.99	0.41	0.55
BHBA (mmol/L)	0.58	0.58	0.56	0.47	0.064	0.13	0.34	0.42
BUN (mg/dL)	15.4	17.5	18.9	19.0	0.97	0.01	0.24	0.22

¹FCM: Fat corrected milk; BW: body weight; BCS: body condition score; BFT: back fat thickness; BHBA: β-hydroxybutyric acid; BUN: Blood urea nitrogen.

The concentrations of BUN increased linearly as BS was replaced for CS (P<0.01). As shown in Table 5, dietary treatment had no effects on plasma concentrations of glucose and BHBA which averaged 64.6 mg/dL and 0.546 mmol/L, respectively, but plasma concentration of BHBA was numerically lower in cows fed CS₀BS₃.

Feeding behavior

Time spent eating (P=0.01) and total chewing time (P=0.04) were quadratically affected with the highest values observed in cows fed CS₃BS₀ or CS₀BS₃ diets (Table 6). The eating time as well as total chewing time per unit of DM, NDF, peNDF, and uNDF_{30h} intakes were also higher in cows fed CS₃BS₀ or CS₀BS₃ diet than cows on other diets. The time spent rumination and rumination time per unit of nutrient intake did not differ as levels of BS increased in the diets. All cows sorted for short (1.18-8 mm) and fine (<1.18 mm) particles and against long (>19 mm) and medium (8-19 mm) particles (Table 7). However, the sorting activity against longer particles was lower in cows fed either CS₃BS₀ or CS₀BS₃ diet.

Discussion

The present study investigated the effects of different

ratios of BS: CS from 3:0 to 0:3 on the productive performance of high producing dairy cows. Both BS and CS were harvested at dough stage of maturity. The stage of harvesting can influence the quality (chemical composition and digestibility) and quantity of the silage materials (Nazli et al., 2019). The NDF content for BS varies widely from 35.3 to 77.3 % (NRC, 2001; Nair et al. 2016). The nutritional characteristics of BS in our study were almost similar to the mean of reported values, showing relatively a medium quality for BS (NRC, 2001, Benchaar et al., 2013).

The optimal corn plant harvesting is recommended at about 30-35 % DM or dent stage with 1/3 to 2/3 milk line to optimize the balance between quality and quantity. However, the timing of harvesting depends on other factors, including the climatic conditions. For example, cool environment or frost can limit the length of the growing season, and thus kernel filling, maturation, and the whole plant DM content (Daynard, 1978). Likewise, corn crop may be cultivated as a second crop in some areas, making a shorter growing season. Chemically, the DM (27.6), NDF (53.2 %), and ADF (30.4) contents of the CS used in the current study were approximately intermediate between the immature (DM = 23.5 %, NDF = 54.1 %, ADF = 34.1 %) and normal (DM = 35.1 %, NDF = 45.0 %, ADF = 28.1 %) corn silages (NRC, 2001).

²L: linear effect, Q: quadratic effect, C: cubic effect.

³ Feed efficiency = (Milk / DMI) and (Corrected milk / DMI).

Table 6. Feeding behavior in lactating cows fed different ratios of corn silage to barley silage

Itom	(Corn silage: Barley silage					<i>P</i> -value ¹		
Item	3:0	2:1	1:2	0:3	SEM	L	Q	C	
Eating									
Min/d	308	277	271	303	18.4	0.63	< 0.01	0.93	
Min/kg DM intake	10.2	8.83	8.70	9.67	0.763	0.37	0.01	0.64	
Min/kg NDF intake	32.6	27.5	27.1	29.1	2.18	0.05	< 0.01	0.17	
Min/kg uNDF _{30h} intake	57.1	49.3	46.8	53.5	4.27	0.17	< 0.01	0.80	
Min/kg peNDF intake	48.9	40.3	39.3	42.9	3.58	0.05	< 0.01	0.19	
Rumination									
Min/d	496	492	484	494	21.3	0.84	0.68	0.84	
Min/kg DM intake	16.2	15.7	15.7	15.8	0.93	0.72	0.63	0.80	
Min/kg NDF intake	52.4	49.0	48.8	47.4	2.78	0.15	0.66	0.28	
Min/kg uNDF _{30h} intake	90.9	87.6	84.2	87.4	5.10	0.39	0.39	0.99	
Min/kg peNDF intake	77.8	71.6	70.8	70.0	4.18	0.11	0.40	0.27	
Total chewing									
Min/d	804	768	755	799	32.6	0.66	0.03	0.82	
Min/kg DM intake	26.4	24.5	24.4	25.5	1.50	0.47	0.07	0.64	
Min/kg NDF intake	84.9	76.4	75.8	76.7	4.42	0.03	0.06	0.11	
Min/kg uNDF _{30h} intake	148	137	131	141	8.1	0.15	0.02	0.89	
Min/kg peNDF intake	126	112	110	113	6.8	0.02	0.03	0.11	
Sorting index									
>19 mm (%)	89.8	93.7	97.7	94.9	3.00	0.03	0.10	0.78	
8-19 mm (%)	98.1	95.6	96.9	98.1	0.58	0.18	< 0.01	0.10	
1.18-8 mm (%)	101.1	101.2	100.8	101.4	0.43	0.78	0.29	0.15	
<1.18 mm (%)	101.2	101.9	100.8	100.8	0.52	0.14	0.31	0.34	

¹L: linear effect, Q: quadratic effect, C: cubic effect.

Nonetheless, CS from the present experiment showed relatively higher content of ADL (5.4 %) than both immature and normal silages (2.5-3.5 %, NRC, 2001; Refat et al. 2017a; Benchaar et al., 2013), but similar to those reported in Nazli et al. (2019). The variations among the results could be explained by several factors, including the plant varieties and climatic conditions. Barley silage had higher concentrations of CP and ammonia-N and pH value than CS. The high protein and ash contents and lower levels of NFC (mainly starch) in BS could be responsible for increased buffering capacity and pH value.

In the current study, DM intake (31 kg/d), NEL intake (50 Mcal/d), milk yield (54 kg/d), and milk yield efficiency (1.74) were typical of high producing dairy cows. These values, as well as milk composition and BW changes, were not affected by feeding different ratios of BS to CS, which contrast with some studies (Refat et al., 2017a; Benchaar et al., 2014). In these experiments, the NDF content of the CS diets was considerably lower than that of BS diets. For example, Benchaar et al. (2014) reported decreases in DM intake by 16 % and milk production by 14 % with increasing proportion of BS which was accompanied with increased NDF concentration (from 32.4 to 37.2 %). Hence, they suggested the de-

creased performance can be related to increased fiber content and its subsequent effect as the BS proportion increased in the diet. Digestibility of NDF is another important parameter among forage types affecting animal performance (Oba and Allen, 1999a). The higher NDF digestibility is associated with an increase in DM intake, and consequently provide more energy for milk production (NRC, 2001). Dado and Allen (1996) fed silages with similar NDF but different NDF digestibility to lactating dairy cows and found significant differences in DM intake and milk yield. In the present experiment, the two silages were different in iNDF and pdNDF contents. However, no differences were found between BS and CS in NDF content (54 % of DM), NDFd₃₀ (42.8 % of NDF), uNDF_{30h} (58.8 % of NDF), NDF digestion rate (1.8 %/h), and TTNDFD (35 % of NDF). Additionally, the apparent NDF digestibility of total diets (44.6 %) did not differ across the treatments. A previous study (Lopes et al., 2015) confirmed that the NDF digestibility coefficient predicted by the in vitro TTNDFD method was a good predictor of in vivo fiber digestion. However, a greater TTNDFD for in situ method vs. in vivo (47.7 vs. 44.6 %) was observed in this study, which may be explained by the higher DM intake as compared to the cows used in Lopes et al. (2015). Together, the lack of a significant effect on milk production or composition, BW gain, or feed efficiency can be attributed to the limited effect of the treatments on NDF digestibility and DM intake. In accordance with our results, DM intake and milk yield (Tudisco et al., 2010; Migliorati et al., 2017; Hosseini et al., 2019) and milk component concentration (Refat et al., 2017a; Migliorati et al., 2017) were not different between CS and BS.

In the current study, the daily intakes of NDF, and iNDF (uNDF_{288h}), uNDF_{30h} increased with increasing BS level in the diet which is likely due to the numerically greater NDF content of the BS diets (~ 1 %) and DM intake (~ 0.75 kg). However, uNDF_{30h} intake, as a percentage of BW, was relatively constant among treatments at approximately 0.87 % of BW (from 0.85 to 0.88 % of BW). Previous experiments also found that different NDF digestibility (Fustini et al., 2017) or forage sources (Kahyani et al., 2019) affected the DM and iNDF intakes, but led to the same intake of uNDF_{24h} or uNDF_{30h}. The filling effect of diets with high or low fiber digestibility could be related to the rate of digestion of pdNDF fraction rather than uNDF_{240h} (Grant and Cotanch (2012). Ferraretto et al. (2015) reported increases in DM intake and milk yield by cows fed CS with higher 30-h in vitro NDF digestibility (brown midrib vs. floury-leafy hybrid). Generally, a negative relationship between uNDF_{30h} and fiber digestibility and pdNDF digestion rate has been reported (Oba and Allen, 1999a; Lopes et al., 2015). These results suggested that intake of uNDF_{30h} (% of BW) may be a better indicator of feed intake in dairy cows over NDF intake which has been suggested to be about 1.25 % of BW (Mertens, 2016). In other words, dairy cows maximize their intake to meet fill limitation which can be better predicted by the uNDF_{30h} intake rather than NDF intake.

In our study, the inclusion of BS linearly decreased DM and OM digestibility by 8 percentage points. Barley silage had similar NDFd30h or uNDF30h, but higher iNDF as compared to CS. The higher amount of iNDF (lower pdNDF) in BS probably decreased the amount of OM available for digestion, which can account for lower DM digestibility. Lopes et al. (2015) showed that for each percentage unit increase in iNDF content, 0.96 percentage unit reduction in TTNDFD can be expected. Lund et al. (2007) and Lopez et al. (2015) observed that pdNDF had higher mean retention time and lower passage rate than iNDF. Consequently, the passage rate of diets with greater BS probably increased because of greater iNDF and lowered digestibility. Furthermore, the lower DM digestibility with increasing BS could be attributed to lower starch content in BS than CS as the starch digestibility is greater (almost twice) than NDF (NRC, 2001). The overall results suggested that uNDF_{30h} was associated more with NDF digestibility and feed intake and iNDF associated with total tract DM digestibility.

The ruminal pH was similar between cows fed CS or BS. However, with increasing BS inclusion in the diets, the content of peNDF and particle size was increased. However, increasing peNDF content may not necessarily result in improved ruminal pH (Beauchemin and Yang, 2005). These authors reported poor correlation (r^2 < 0.13) between ruminal pH and dietary fiber measured as NDF, forage NDF or peNDF. It was concluded that factors other than dietary NDF such as DM intake, diet fermentability, and feeding management practices also influence ruminal pH (Zebeli et al., 2012). The similar pH value between the treatments is partly attributable to the fact that the DM intake and ruminating time were the same across the treatments. Previous studies reported that ruminal ammonia concentration increased when BS replaced CS in the cow diet (Hosseini et al., 2019; Benchaar et al., 2014). The availability of fermentable carbohydrates strongly influences the utilization of ammonia in the rumen (Hristov et al., 2005). The CS had higher starch than BS; this may improve the microbial utilization of dietary N and decrease the ruminal ammonia concentration. The increase in ruminal ammonia-N concentration with feeding BS may be explained by a lower supply of energy for ruminal microbes due to its greater iNDF coupled with a lower OM digestibility, as reflected by the increase in BUN. Refat et al. (2017b) reported diet containing CS had higher microbial protein production than diet containing BS; suggesting that CS has higher potential to provide adequate amounts of energy for the synthesis of microbial N. Concentrations of glucose BHBA were consistent between cows fed BS or CS. The lack of responses in glucose or BHBA could be attributed to similar DM intake and milk yield across the treatments.

It was expected that chewing time increases with increasing dietary substitution of BS for CS because of increased dietary mean particle size (6 %), peNDF_{1.18} (9 %), and iNDF (16 %). However, all diets contained the same amount of NDF (32.3 %), forage NDF (19 %), and uNDF_{30h} (18.1 %). Total chewing time per DM or uNDF_{30h} intake was quadratically decreased as the level of BS increased in the diet. The longer chewing time in cows fed CS₃BS₀ or CS₀BS₃ diet was corresponded to eating time. In a previous study (Hosseini et al., 2018), a numerically lower eating time but greater rumination time was observed in cows fed BS as compared to cows fed CS. Lack of ef-

fect of treatment on rumination activity in this experiment may be attributed to the limited effect on the DM intake and the higher iNDF in BS. The greater iNDF, with a greater passage rate than pdNDF, may be the reason for similar rumination time and DM intake among cow groups fed BS and CS. Regardless of treatment, all cows sorted against long (>19 mm) and medium (8-19 mm) particles, but cows on CS₃BS₀ or CS₀BS₃ diet had greater sorting for medium particles and against long particles. These results may contrast with the common view that feeding long particles has a greater effect on eating time, whereas intake of medium particles (4-19 mm) affects rumination time (Beauchemin et al., 2018). We did not find any explanation why cows fed the CS₃BS₀ or CS₀BS₃ diets had higher eating time than the other two treatments. Intake of peNDF is the main contributing factor to rumination time (Beauchemin et al., 2018; Zebeli et al., 2012).

Conclusions

Fiber digestibility is an important parameter of forage quality that has a profound impact on various aspects of animal responses, including ruminal fill and DM intake, diet digestibility, feeding behavior, and milk yield in dairy cows. In this work, CS and BS had similar predicted TTNDFD and uNDF_{30h} that could be indicators of rumen fill and fiber digestibility, and potential feed intake. Under these conditions, partial or total substitution of CS with BS had no negative impact on chewing time, apparent digestibility of NDF, DM intake, and milk yield of lactating cows. However, feeding BS diet lowered DM digestibility which can be attributed to the higher BS iNDF content. Our results suggested that CS can be substituted with BS in the diet of high producing dairy cows when fiber digestibility is similar between the silages.

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