Effects of fiber source on apparent digestibility and ruminal fermentation parameters in sheep fed high-concentrate diets

M. Mohammadi, F. Ahmadi, M.J. Zamiri*

Department of Animal Science, College of Agriculture, Shiraz University, Shiraz, Iran * Corresponding author, E-mail address: zamiri@shirazu.ac.ir

Abstract Sixteen rams (mean age: 13 mo; mean live weight: 40.0 ± 2.4 kg) were randomly allotted to four dietary treatments in a completely randomized design (4 rams per treatment). Diets (dry matter basis) contained 65% concentrate and 35% alfalfa hay (control diet, T1), 35% wheat straw (T2), 35% barley straw (T3) or 35% maize straw (T4). Total-tract apparent digestibility for dry matter, organic matter, neutral detergent fiber (ADF), acid detergent fiber (ADF), and crude fat was not affected by the diet (P > 0.05). Among straws, maize straw had the highest crude protein digestibility of 63%, compared with wheat straw (48%) and barley straw (54%). Greater nitrogen balance was recorded for diets containing alfalfa hay and maize straw. Gas production volume after 72 h incubation, was higher in the diet containing alfalfa hay or maize straw compared to that containing barley or wheat straw. Ruminal fluid pH and NH₃-N were not affected by straw type. In conclusion, the diet containing maize straw was superior to diets containing either wheat or barley straw in terms of crude protein digestibility, nitrogen balance, and *in vitro* ruminal fermentation parameters. This associative effect of fiber type in high-concentrate diets could be important in practical sheep feeding, as it may affect the animal performance.

Keywords: straw, digestibility, forage: concentrate ratio, sheep

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Introduction

Global demand for food of animal origin is expected to increase substantially (Haines et al., 2009), hence improvement in animal production systems is needed to address the future food crisis. Cereal straws are inexpensive and potentially good sources of energy for ruminants, and both in dry or winter times and cropping seasons, they can constitute a major proportion of the ruminant diet (Ghasemi et al., 2014), however, due to low availability of structural carbohydrates and insufficient contents of nitrogen, minerals and vitamins, they cannot efficiently be utilized by the ruminant animal (Leng, 1990; Izadifard and Zamiri, 2007;Mahesh and Mohini, 2013).

Nutrient utilization in straw-based diets can be increased by concentrate supplementation, primarily by supplying fermentable carbohydrates and nitrogen (N) to ruminal microorganisms (Molina-Alcaide et al., 2000, Shem et al., 2003, Haddad and Husein, 2004, Tessema and Baars, 2004). Positive N balance is usually achieved in animals fed high-concentrate diets (Tripathi et al., 2007) by reducing nutrient loss to the environment (Bach et al., 2005). Concentrate-based diets cause greater weight gain (McDonald et al., 2002) and minim-

ize energy and thus lead to enhanced efficiency of utilization of dietary energy for body weight gain (Mandebvu and Galbraith, 1999). However, high-concentrate diets promote acidosis in ruminants (Owens et al., 1998) and disturb the normal function of the rumen; this necessitates inclusion of a some fiber to stimulate rumination and salivary secretion (Kawas et al., 1991, Van Soest, 1994). High-concentrate diets may also negatively affect fiber digestion and increase the lag time of fiber digestion (Grant, 1994). However, fiber digestibility is highly dependent on forage quality; the low-quality forages are more prone to negative associative effects when high-concentrate feeds are incorporated in the diet than are high-quality forages (Cerrillo et al., 1999). The positive effect of the concentrate supplements on digestion of forages is achieved by provision of nutrients such as N and phosphorus which may be deficient in some forages and fibrous agricultural byproducts (Dixon and Stockdale, 1999).

Lamb meat is the primary source of red meat in Iran (Papi et al., 2011) and profitability depends greatly on minimizing production costs. Improvements in feed efficiency without negatively affecting the animal perfor-

mance help the intensive sheep-rearing units to operate on a higher profit margin (Snowder and Van Vleck, 2003, Haddad and Ata, 2009).

This study was conducted to compare the effects of barley straw, wheat straw, and maize straw with alfalfa hay on apparent total-tract nutrient digestibility and selected ruminal parameters in rams fed a high-concentrate diet. This information may be helpful in better understanding of the interaction effect between forages and concentrate which is important in terms of efficiency of feed utilization.

Materials and methods

Animal housing and experimental diets

The experiment was carried out at the Animal Research

Station, College of Agriculture, Shiraz University, Shiraz, Iran. Sixteen 13-month-old rams (mean live weight: 40.0 ± 2.4 kg; mean \pm SD) were housed in individual metabolic crates (100×100 cm). Sun-dried straws, provided from the nearby fields, were chopped into 2 to 3 cm long pieces and mixed with the concentrate as a total mixed diet. Ingredients and chemical composition of the experimental diets, formulated according to the NRC recommendations (NRC, 2007), are presented in Table 1. Diets (dry matter basis) contained 65% concentrate in common and 35% alfalfa hay (control diet, T1) or, 35% wheat straw (T2) or, 35% barley straw (T3) or 35% maize straw (T4). Daily feed (1700 g consisting of 1100 g concentrate and 600 g straw) was fed as ad libitum twice a day (08:00 and 16:00). Diets were offered for 21

Table 1. Ingredients and chemical composition of the diets (% of DM unless otherwise indicated)

	Diets ¹			
-	T1	T2	Т3	T4
Ingredients				
Alfalfa hay	35			_
Wheat straw	_	35	_	_
Barley straw			35	
Maize straw	_	_	_	35
Barley grain	47.5	44.0	44.0	43.8
Cottonseed meal	7.0	9.0	9.0	9.0
Wheat bran	9.0	8.0	8.0	10
Urea	0.0	0.5	0.1	0.11
Protected fat	0.0	1.5	1.9	0.1
Calcium carbonate	0.5	1.0	1.0	1.0
Common salt (NaCl)	0.5	0.5	0.5	0.5
Vitamin-mineral premix ²	0.5	0.5	0.5	0.5
Chemical composition				
Ash^3	6.8	8.2	9.9	7.0
Ether extract (EE) ³	1.6	2.4	4.2	2.1
Crude protein (CP) ³	13.0	12.1	12.0	12.2
Calcium	0.53	0.45	0.50	0.52
Phosphorus	0.44	0.39	0.40	0.46
NDF^3	36.4	43.6	42.5	40.6
ADF^3	22.1	26.0	23.9	21.3
NFC^4	42.2	33.7	31.4	38.1
ME ⁵ , Mcal/kg	2.48	2.41	2.47	2.46

¹ Diets contained (DM basis): 65% concentrate with 35% alfalfa hay (T1), wheat straw (T2), barley straw (T3) or maize straw (T4).

² Vitamin-mineral premix contained per 100 g: 500,000 IU vitamin A; 10,000 IU vitamin D₃; 100 mg vitamin E; 180 mg Ca; 90 mg P; 2000 mg Mn; 3000 mg Fe; 300 mg Cu; 100 mg Co; 3000 mg Zn; 55 g Na; 19 g Mg

³ Based on chemical analysis of individual feedstuffs

 $^{^4}$ NFC = non-fiber carbohydrates calculated from ingredients as NFC = 100 - (NDF + CP + EE + ash).

⁵ Metabolizable energy; based on tabulated data (NRC, 2007).

Mohammadi et al.

d including 10 d for dietary adaptation and 11 d for sample collection. Fresh clean water was freely available throughout the experiment.

Sampling procedure

Prior to the morning feeding, the orts were weighed (using a digital balance readable to 5.0 g), and pooled for each animal; the offered feed was 15% in excess of the previous day's intake (Forbes, 2007). After 11-day total collection of feces (feces was collected prior to the morning feeding), the fecal samples (10% of the total excretion) were pooled to form a composited sample for each animal by period, and then stored inside a ziplocked plastic bag at -20°C. The fecal sample (10 g) was dried to a constant weight in a forced-air oven at 55°C for 48 h, and then sieved through a 1-mm screen for subsequent chemical analysis. The apparent nutrient digestibility was calculated as the nutrient intake not recovered in feces (McDonald et al., 2010). For urine sampling, a 100-mL urine sample was transferred into a plastic container containing 10% (v/v) sulfuric acid (pH 3.0) to minimize ammonia loss and frozen at -20°C pending analysis. Ruminal fluid was collected on three occasions: at the onset of the adaptation period, and at the beginning and end of sample collection; the collections were made prior to the morning feeding (t = 0) and at 2 and 4 h post-feeding using an electric vacuum pump. Immediately after sampling, ruminal fluid pH was determined using a pH meter (M/s Jenway Model 3510, Camlab, Cambridge, UK). Ruminal contents were squeezed through 4 layers of cheesecloth and rumen fluid (10 mL) was then placed into bottles containing 2 mL of 25% metaphosphoric acid and stored (-20°C) until analyzed for ammonia. N balance was calculated by subtraction of the amount of average daily N intake from the average daily N excreted in the feces and urine (McDonald et al., 2010).

Chemical analyses

For chemical analysis, the samples were ground and milled through a 1-mm screen. NDF and ADF concentrations were determined sequentially using thermo-stable alpha-amylase and sodium sulfite (Van Soest et al., 1991). Crude protein (CP, N \times 6.25, method No. 984.13), ether extract (EE, method No. 954.02), dry matter (DM, method No. 930.15), and ash (method No. 942.05) contents were measured according to Association of Official Analytical Chemists (1990). Organic matter (OM) content was calculated as the difference between sample DM weight and ash content.

Ruminal NH₃-N

Samples were thawed overnight, and then centrifuged at 12,000 g for 20 min at 4°C to obtain a clear supernatant. The supernatant was analyzed for rumen ammonia with a phenol-hypochlorite reaction method (Broderick and Kang, 1980).

Table 2. Comparative effect of straw type on nutrient digestibility, nitrogen balance and *in vitro* gas production parameters in rams fed a high-concentrate diet

	Diets ¹			SEM	<i>P</i> -value	
•	T1	T2	Т3	T4	SEM	r-value
Apparent digestibility (%)						
Dry matter	66.4	57.3	60.8	64.9	3.71	0.351
Organic matter	69.3	64.7	66.4	68.2	1.88	0.331
Crude protein	63.4a	48.4^{b}	54.3ab	62.8a	2.82	0.007
Neutral detergent fiber	61.4	57.2	53.3	57.3	4.24	0.103
Acid detergent fiber	50.4	45.1	41.5	45.8	5.26	0.335
Ether extract	60.0	46.6	51.3	54.1	3.34	0.083
Nitrogen balance (g/d)	11.3 ^a	5.2 ^b	2.8^{b}	10.4 ^a	4.49	< 0.001
GP_{72}^2	307.4^{a}	283.6 ^b	291.7 ^b	305.6a	3.86	0.050
Rate constant $(h^{-1})^3$	0.07^{a}	0.03^{d}	0.04^{c}	0.05^{b}	0.001	0.050
Lag time (h)	0.06^{b}	0.40^{a}	0.20^{ab}	0.10^{b}	0.056	0.050

¹T1 = 65% concentrate + 35% alfalfa hay; T2 = 65% concentrate + 35% wheat straw; T3 = 65% concentrate + 35% barley straw; T4 = 65% concentrate + 35% maize straw

² Cumulative gas production (mL/g DM) at the end of 72 h in vitro incubation

³ Determined by a first-order exponential model proposed by Ørskov and McDonald (1979)

a-d Within each row, means with common superscript (s) are not different (P > 0.05; Tukey's test).

Table 3. Effect of straw type on ruminal fluid pH at the start and end of experiment in rams fed a high concentrate diet

Diets ¹	Start	End		
T1	A7.06a	A6.89a		
T2	^B 6.73 ^a	$^{A}6.89^{a}$		
T3	$^{\mathrm{B}}6.74^{\mathrm{a}}$	A6.94a		
T4	$^{\mathrm{AB}}6.87^{\mathrm{a}}$	A6.91a		

¹ Diets contained (DM basis): 65% concentrate with 35% alfalfa hay (T1), wheat straw (T2), barley straw (T3) or maize straw (T4).

Cumulative gas production

Two rumen-fistulated non-lactating Holstein cows were fed a diet of alfalfa hay (30%), wheat straw (30%) and concentrate mix (40%) containing a mineral/vitamin supplement. Rumen liquor was collected before the morning feeding. Procedures for preparation of rumen fluid and artificial saliva were those described by Ahmadi et al. (2013). Volume of gas in each volume-calibrated serum flask was manually measured using a water displacement apparatus (Fedorak and Hrudey, 1983).

Statistical Analysis

The experiment was performed as a completely randomized design. Data were analyzed using the GLM procedure (SAS, 2003) according to the following model:

$$Y_{ij} = \mu + T_i + e_{ij}$$

where, Y_{ij} , μ , T_i , and e_{ij} represent the measured value for each observation, overall mean, treatment effect, and the random residual error, respectively. Ruminal NH₃-N and pH data were analyzed using PROC MIXED for repeatedly-measured observations. Mean separation was performed using the Tukey's test (P < 0.05).

Results

Ingredients and chemical composition of the experimental diets are presented in Table 1. The NDF content was the highest for the wheat straw-containing diet followed by that containing barley straw and then in those containing maize straw and alfalfa hay, whereas that of NFC was highest for the alfalfa hay-containing diet followed by that containing maize straw and then in those containing wheat and barley straw (Table 1). Protein content of T1 (13.0%) was slightly higher than that of other diets (12.0 to 12.2 %).

Apparent digestibility and *in vitro* gas production data are shown in Table 2. Total-tract apparent digestibility of DM and OM was not affected by the diet (P > 0.05). Apparent CP digestibility was higher (P < 0.05) for maize straw (63%) than for wheat straw (48%), with barley straw having an intermediate value (54%). Appa-

rent digestibility of ADF and NDF was not affected by the diet (P > 0.05). Crude fat (ether extract) digestibility was lower for wheat straw diet (46%) compared with barley straw (51%) and maize straw (54%) diets (P = 0.0831). *In vitro* gas production and N balance were greater for the diet containing maize straw compared with wheat and barley straw (P > 0.05).

The interaction effect between diets and day of sampling was the only factor which significantly affected the ruminal fluid pH values (Table 3). Ruminal fluid pH was higher for T1 at the beginning of the experiment but at the end of the experiment, no significant differences were found between the diets. None of the diets showed any significant difference in pH values between the initial and last day of ruminal fluid collection.

Changes in ruminal NH₃-N concentration at various experimental periods [*i.e.* start of adaptation period (phase 1), beginning of data collection (phase 2), and the end of data collection (phase 3)] are presented in Table 4. At the beginning of the adaptation period, the difference in ruminal NH₃-N concentration was not significant amongst diets (P > 0.05). At phases 2 and 3, diet T1 resulted in a slightly higher ruminal NH₃-N concentration compared with other diets. No significant effect of the day of sampling on NH₃-N was found in the diets that contained straw.

Discussion

Higher *in vitro* ruminal digestibility (in terms of 72-h cumulative gas production and gas production rate constant) of T1 and T4 diets was most likely due to their lower NDF content, and at the same time their higher NFC content, since the latter is considered as the least digestible component in forages and the former is characterized by its high inherent digestibility (Falls, 2011). There is a general agreement that increased levels of concentrate in the ruminant diet are accompanied by increases in DM and OM digestibility (Molina-Alcaide et al., 2000; Fimbres et al., 2002). In Omani growing lambs, increasing the dietary energy density from low to

A.a within each row (lowercase letter) or column (uppercase letter), means with common superscript (s) are not different (P > 0.05; Tukey's test). Overall SEM = 0.075.

Mohammadi et al.

Table 4. The effect of straw type on ruminal fluid NH_3 -N concentration (mg/100 mL) in rams fed a high-concentrate diet

	8		
Diets ¹	Start of adaptation period	Start of experiment	End of experiment
T1	A15.12b	A16.47a	A16.58a
T2	^A 14.24 ^a	B14.30a	B13.85a
T3	A14.22a	$^{\rm B}14.24^{\rm a}$	B14.73a
T4	A15.10 ^a	B14.19a	^B 14.63 ^a

¹ Diets contained (DM basis): 65% concentrate with 35% alfalfa hay (T1), wheat straw (T2), barley straw (T3) or maize straw (T4).

high, resulted in an increase in DM digestibility from 66.8 to 73.3% (Mahgoub et al., 2000). However, negative or positive associative effects, where forages are incorporated in high-concentrate diets, depend greatly on digestibility of the fibrous components of the forage (Dixon and Stockdale, 1999). Cantalapiedra-Hijar et al. (2009) investigated the effects of forage-to-concentrate ratio and two forages of different quality (grass hay vs. alfalfa hay) on ruminal fermentation in goats. High-concentrate diets tended to have higher CP digestibility, N retention, and ruminal NH₃-N concentration in animals fed diets based on grass hay (higher-quality forage). Shifting the forage- to- concentrate ratio from 70:30 to 30:70 was more beneficial to digestibility in grass hay which had a better quality in terms of less structural carbohydrates compared with alfalfa hay (ADF content of 273 and 320 g/kg of fresh matter for grass hay and alfalfa hay, respectively). It was also reported that N was retained more efficiently in goats fed diets based on grass hay when the concentrate level increased. These results emphasize the positive associative effect of feed when high-concentrate diets are incorporated with highquality forage diets (Cerrillo et al., 1999).

A ruminal pH below 6.2 is reported to depress fiber digestion by inhibiting the growth of cellulolytic bacteria (Grant and Mertens, 1992), which is likely the result of a reduction in rumination and saliva secretion (Van Soest, 1994). However, in the present study, the ruminal pH was not lower than 6.2 at any time point during the experiment, suggesting that the level of concentrate (65%) in the experimental diets did not exert any detrimental effect on fiber digestion due to pH drop. An interesting result of Tripathi's study on the effect of different levels of concentrate in weaner lambs was that the higher concentrate level (80% of total DM) did not induce acidosis, where the ruminal pH remained above 6.4 during post-feeding hours (Tripathi et al., 2007), emphasizing the fact that feeding high-concentrate diets do not necessarily result in low ruminal pH.

Ruminal NH₃-N is a crucial nutrient which is necessary

for efficient rumen fermentation, with ammonia-N being being used as a N source to improve rumen ecology (Wanapat and Pimpa, 1999). At high NH₃-N concentrations, various types of bacteria, protozoa, and fungi may occur, thus leading to better functioning of the rumen. For example, a study on swamp buffalo showed that ruminal NH₃-N concentrations in the range of 13.6–17.6 mg/dL improved rumen ecology, which was reflected in an improvement in digestibility and intake of rice straw (Wanapat and Pimpa, 1999).

Increasing the concentrate portion in a diet based on grass hay but not in an alfalfa hay diet that contained more structural carbohydrates, increased ruminal NH₃-N concentration (P<0.05) in Granadina goats (Cantalapiedra-Hijar et al., 2009). In the current experiment, ruminal NH₃-N concentration in all dietary treatments was substantially above the level suggested (5 mg/100 mL) to maximize microbial protein synthesis (Satter and Slyter, 1974). High-concentrate diets promote the incorporation of ruminal NH₃-N into microbial protein, which is reflected in higher N balance (Sultan et al., 1992). The higher N balance means the higher protein digestibility and thus reduced N excretion, which may be an indication of improved microbial protein efficiency. Our data (Table 2) suggested that absorbed N may be more efficiently retained in the case of T1 (alfalfa hay) and T4 (maize straw) diets compared with T2 (wheat straw) and T3 (barley straw) diets. This is a result of higher ruminal fermentability of T1 and T4 (Table 2), leading to higher N retention as compared with other dietary treatments. Increasing the level of concentrate fed (15 or 25 g kg⁻¹ body weight, or ad libitum) increased N retention in weaner lambs, highlighting the important role of concentrate level in N balance (Tripathi et al., 2007). Our results indicated that interaction effect between the forage and concentrate was dependent on the forage quality; the level of concentrate used in the present experiment more positively affected the higher-quality straw (less structural carbohydrates) in terms of higher N balance.

 $^{^{}A.a}$ within each row (lowercase letter) or column (uppercase letter), means with common superscript (s) are not different (P > 0.05; Tukey's test). Overall SEM = 0.301.

Conclusion

In a diet containing 65% concentrate mix and 35% straw, maize straw was comparable to alfalfa hay, and generally superior to wheat and barley straw in terms of apparent nutrient digestibility, *in vitro* gas production and N balance. Maize straw, where available, can substitute alfalfa hay when the latter is in short supply or when its use is prohibitive because of high price. This also results in a more efficient use of crop residues in places where good quality forges are expensive or less abundant.

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Mohammadi et al.

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م. محمدی، ف. احمدی و م. ج. ضمیری ဳ

نویسنده مسئول، یست الکترونیک: zamiri@shirazu.ac.ir

چکیده در یک طرح کاملاً تصادفی، شانزده قوچ (۱۳ ماهه و با میانگین وزنی و انحراف معیار ۴۰/±۴۰/۰ کیلوگرم) با چهار جیره غذایی تغذیه شدند. جیرهها (برپایه ماده خشک) دارای ۶۵ درصد کنسانتره همراه با ۳۵ درصد یونجه خشک (جیره شاهد)، ۳۵ درصد کاه گذرم، ۳۵ درصد کاه جو، یا ۳۵ درصد کاه ذرت بودند. گوارشپذیری ظاهری ماده خشک، ماده آلی، فیبر های محلول در شویندههای اسیدی یا ختثی، و چربی خام (عصاره اتری) جیرهها تغییرات معنی داری نشان نداد (۲۰/۰۵). ضریب گوارشپذیری پروتین خام جیره دارای کاه ذرت (۶۳ درصد) بالاتر از این ضریب در کاه گندم (۴۸ درصد) و کاه جو (۵۴ درصد) بود (۲۰/۰۵).. حجم گاز تولیدی پس از ۲۷ ساعت در شرایط برون تنی، برای جیرههای دارای یونجه خشک یا کاه ذرت بالاتر از تولید گاز برای جیره دارای کاه گندم یا کاه جو بود. منبع فیبر (کاه) تأثیر معنی داری بر اسیدیته و غلظت نیتروژن آمونیاکی در مایع شکمبه نداشت. یافتهها نشان دادند که جیره دارای کاه درت تا در حنبه گوارشپذیری ظاهری پروتین خام، تعادل نیتروژن و فرآیندهای تخمیری شکمبه برتر از جیرههای دارای کاه گندم یا کاه جو بود. این اثر همراهی فیبر خوراک در یک جیره پر کنسانتره می تواند از دیدگاه تغذیه کاربردی اهمیت داشته باشد، زیرا ممکن است بر عملکرد اثر بگذارد.