Feeding Value of Ammoniated Rice Straw Supplemented with Rice Bran in Sheep: II. *In Situ* Rumen Degradation of Untreated and Ammonia Treated Rice Straw

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ABSTRACT : The effect of ammonia treatment and rice bran supplementation on the *in situ* rumen degradation of rice straw was determined using three Japanese Corriedale wethers fitted with permanent rumen cannula. About 4 g samples of diets containing 100% untreated rice straw (URS); 100% ammonia treated rice straw (ARS); 65% URS+30% rice bran (RB)+5% soybean meal (SBM) (T1); and 85% ARS+15% RB (T2) were incubated at 0, 4, 8, 16, 24, 48, 72, and 96 hours in the rumen of sheep to measure dry matter (DM), crude protein (CP) and neutral detergent fiber (NDF) degradability. The DM disappearance of ARS based diets were about 20% higher than that of URS based diets. Rice bran supplementation improved DM disappearance of URS but not on ammoniated straw. Degradation parameters showed that ammoniation increased rate (c) of straw degradation resulting to higher DM and fiber degradability but RB supplementation did not. ARS gave similar DM and CP solubility and effective rumen degradability (ED) with that of the supplemented groups indicating that ammoniation alone can give the same effect on rumen degradability of sheep receiving low quality roughage. All degradation parameters for NDF were consistently higher in ARS based-diets indicating improved fiber solubility. Rice bran supplementation did not affect degradation characteristics of the diets except on soluble DM and CP fraction (A) of URS but not on ARS. (***Asian-Aus. J. Anim. Sci. 2000. Vol. 13, No. 7 : 906-912***)

Key Words : *In Situ* Degradation, Untreated Rice Straw (URS), Ammoniated Rice Straw (ARS), Rice Bran (RB), Supplementation

INTRODUCTION

The use of cereal straw as feeds for ruminants is characterized by low protein content and high degree of lignification. Several chemical treatments have been tested to delignify or disrupt the lignin-carbohydrate complex present in cereal straw. Among these, ammoniation is the most appropriate because it increase nitrogen (N) content and cellulose degradability of treated straw (Sundstol, 1984; Zorilla-Rios et al., 1985). However, ammoniation alone has not contributed much in meeting nutrient needs of ruminants beyond maintenance level even with increased intake and ruminal degradability (Castrillo et al., 1994; Fike et al., 1995). Thus, there is a need for concentrate supplementation in straw-based diets to support higher levels of production (Castrillo et al., 1994).

Rice bran contains easily fermentable sugars that could provide energy for rumen microbes. However, it has adverse effects on fiber degradability which can negate the benefits that could be derived from incorporation. This is usually associated with low ruminal pH and lactic acidosis (Nocek and Russell, 1988) that tend to reduce activity of fibriolytic bacteria in favor of the amylolytic organism (Maczulak et al., 1981; Zhao et al., 1996a). The minimum level of inclusion which causes this negative effect and the suitability of certain combination (Mgheni et al., 1994) when given as supplement to either untreated or ammoniated straw is not totally clear (Fondevila et al., 1994).

The objective of this study is to determine the effect of ammoniation and rice bran supplementation on the *in situ* ruminal disappearance and degradation characteristics of straw based-diets.

MATERIALS AND METHODS

Rice straw treatment

About 5 kg of chopped rice straw (2-3 cm length) were placed in polyethylene sacks. Air was removed from each sack, then a 25% ammonia solution was infused at the rate of 3 g N/kg DM (Sundstol, 1984) using a 10 liter capacity knapsack sprayer. During spraying, the nozzle was moved in different directions to ensure uniform distribution of the ammonia solution. The bags were then tightly secured with a rope and incubated at room temperature of about 28℃. Weekly turning of the bags was done to avoid excess ammonia solution from sticking in one side of the sack. After 30 days of curing, treated straw was sun-dried for 2-3 days to ensure evaporation of excess ammonia.

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Table 1. Chemical composition of feedstuffs and experimental diets (% DM)

Nutrients	Rice bran	Soybean meal	URS	ARS	T_1	T_2
Dry matter	86.48	87.66	88.87	88.16	89.36	87.84
Organic matter	89.68	91.76	77.93	81.65	82.52	82.76
Crude ash	10.31	8.24	22.06	18.35	17.48	17.23
Crude protein	12.5	45.56	4.0	8.93	9.12	9.43
NDF	21.69	11.23	63.96	60.68	49.86	54.31
ADF	9.37	9.00	40.74	39.27	31.00	37.39
Hemicellulose	12.32	2.23	23.22	21.41	18.86	16.92
Cellulose	6.70	8.70	35.23	35.09	25.88	33.20
Lignin	2.67	0.30	5.51	4.59	5.12	4.19
Silica	0.12	0.23	15.89	10.96	9.86	10.49

URS=Untreated rice straw; ARS=Ammonia treated rice straw.
T_1=65% URS+30% RB+5% SBM. T_2=85% ARS+15% RB.

Experimental animals and diets

Three Japanese Corriedale wethers (approximately 3 years old) with an average body weight of 32 kg and fitted with permanent rumen cannula were kept individually in metabolism crates. The animals were fed at maintenance level, giving 15% allowance for refusal with diet predominantly made up of either URS or ARS supplemented with RB. The amount of RB inclusion was adjusted to meet maintenance energy requirement (NRC, 1985). Feeds were offered in two equal portions at 09:00 and 17:00 h, while water was always made available. A 10-day adaptation period was allowed prior to incubation of the samples.

***In situ* degradation**

The ruminal degradability of the following diets; 100% URS (URS); 100% ARS (ARS); 65% URS+30% RB+5% SBM (T1) and 85% ARS+15% RB (T2) were determined using the nylon bag technique as described by Ørskov and Ryle (1985). Prior to incubation, samples were air dried, and ground through a Wiley mill with a 2 mm sieve. The chemical composition of the feedstuffs and diets used is presented in table 1.

Nylon bags (internal dimension of 8×12 cm; pore size of 47 μm) containing about 4 g air-dried samples were incubated in duplicate in the rumen for 4, 8, 16, 24, 48, 72, and 96 hours. There were 6 replicates for each diet (2 bags per sample x 3 animals). After incubation, bags with residue were taken out of the rumen, dipped immediately into cold water to stop microbial activity, then rinsed with tap water to remove the adhering feed particles outside the bags. Thereafter, the bags were machine washed for 15 minutes without spinning. Three additional bags containing 4 g samples were set aside for 0-h incubation (water soluble fraction=A). All residual samples were dried at 60℃ for 48 h and stored for further chemical analysis. The residues were ground to pass through 1 mm sieve before DM, CP and NDF were determined.

Residual DM, CP and NDF values were fitted to the NEWAY computer program (X. B. Chen, 1995) to determine degradation characteristics of the diets based on the models of Ørskov and McDonald (1979). The model describes the degradation curve as:

$$p=a+b(1-e^{-ct})$$

where: p=degradation after t=time (h); a=soluble or highly degradable fraction; b=slowly degradable fraction which disappears at a constant fractional rate (c); c=degradation rate (per h).

The slowly degradable fraction (b) was re-estimated as B=(a+b)-A, where A=actual soluble fraction (washing loss), as proposed by Ørskov and Ryle (1990).

Chemical analysis

The DM of the feed and rumen undegraded samples were analyzed following the AOAC (1984) procedure, while the CP contents of the samples was determined by Kjeldahl method. Neutral detergent fiber was analyzed following the method of Goering and van Soest (1970).

Statistical analysis

Analysis of variance (ANOVA) was conducted using the General Linear Model (GLM) procedure of Statistica for WindowsTM Released 4.3, (StatSoft, Inc., Tulsa, OK., 1993). Mean comparison was done using Duncans Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

***In situ* DM disappearance**

The DM disappearance of the various diets at different incubation time is presented in figure 1. Dry

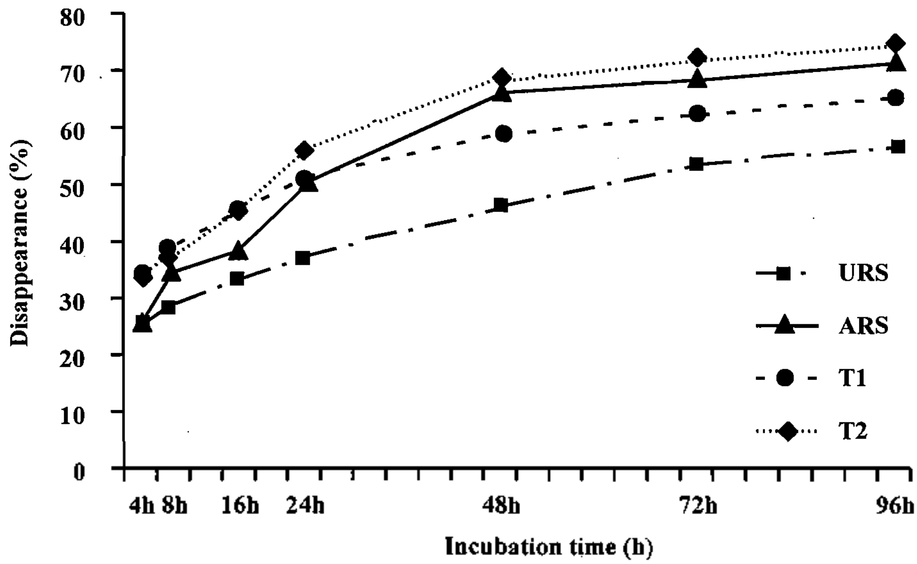

Figure 1. *In situ* dry matter disappearance at different incubation time of URS and ARS based-diets supplemented with RB in sheep

matter disappearance of ARS based-diets was consistently higher than URS based-diets, reaching asymptotic level as incubation time progresses. Ammoniation increased DM disappearance of rice straw by almost 20% after 48 h of incubation. The same improvement was observed by Chiquette et al. (1992) when low quality hay was treated with either ammonia or urea. The degradation at 48 h in this experiment was very close to the mean retention time for low quality feeds in the rumen and to the *in vivo* digestibility (Ørskov and Rlye, 1985). The higher disappearance in ARS based-diets suggests that straw has the tendency to undergo a greater degree of particle disintegration (Van Soest et al., 1984) which provided better adhesion sites for microbial attachment and activity (Grenet and Barry, 1990).

The inclusion of RB improved the degradability of URS based-diets (T1), and provided similar disappearance value with that of ARS and T2. About 50% of DM in ARS, T1 and T2 were degraded after 24 h of microbial rumen activity. The insignificant variation among these diets suggests that treating rice straw with ammonia would give the same DM disappearance with that of supplemented diets. However, during the later stage of incubation, from 48 to 96 h, the ARS based diets had consistently higher DM disappearance than the URS based-diets even with RB supplementation.

Degradation characteristics

Table 2 shows the average straw degradation parameters after fitting the individual DM, CP and NDF disappearance data to the exponential model of Ørskov and McDonald (1979). Ammonia treatment resulted in higher degradability of straw diets. With slowly degradable fraction (B) in ARS based-diets, potential degradability (A+B) was enhanced. Result was similar with those obtained by Fondevila et al. (1994), on barley straw treated with ammonia. They reported an improvement of 149 g/kg degradable DM (A+B) from the straw due to higher B value as a result of ammonia treatment. The rate (c) of straw degradation was also improved by the treatment. While this result was in contradiction with previous reports of Dryden and Kempton (1983), Zorilla-Rios et al. (1985) and Fondevila et al. (1994), it is consistent with the results of Ibrahim et al. (1989), Shen et al. (1998) and Fondevila et al. (1994). They reported that the potential degradability of straw increased due to higher B value as a result of ammonia treatment. Thus, the major effect of ammonia treatment in modifying rumen degradation of straw was on degradable DM (B) fraction (Dryden and Leng, 1988; Fondevila et al., 1993) relative to the improved degradability of the whole cell wall components which accounts to 60% (table 1) of the straw diet. Moreover, Robinson and Kennelly (1988) had the same conclusion in explaining the role of ammoniation on the increased ruminal degradability of high moisture barley in dairy cows.

Ammonia treatment improved the effective degradability (ED) of straw from 43% to 54% at fractional rate of 0.02 and from 34% to 42% at fractional rate of 0.05 per h. The DM degradability measured at particulate passing rate of 0.02 per h is almost similar to the 42% for URS and the 55% for

ARS reported by Mgheni et al. (1994) on fish meal supplemented and urea treated rice straw in sheep. The higher DM degradability of ARS based-diets supports the previous observations that ammoniation increased digestibility (Dryden and Kempton, 1983; Han et al., 1989) and rumen degradability of low quality roughage (Ibrahim et al., 1989; Zorilla-Rios et al., 1985).

Rice bran supplementation of URS and ARS promoted higher ($p<0.01$) soluble DM (A). Rumen degradable DM (B), however, was decreased significantly for URS, ARS and T1 but not between ARS and T2. This result confirms the observation that highly fermentable carbohydrates from cereal grains (Castrillo et al., 1996; Fondevila et al., 1996) or their by-products, such as rice bran (Forster et al., 1993) adversely affect rumen degradability of poor quality roughage materials. The depression could be due to reduced microbial activity with RB supplementation (Zhao et al., 1996) or preferential action of rumen microbes (Devendra and Lewis, 1974). Conversely, the adverse effect of RB inclusion on insoluble but degradable DM fraction was not observed with ARS diets. However, the potential DM degradability (A+B) and rate (c) of degradation was not significantly different between ARS and T2. The improvement in the quality of rice straw brought about by ammoniation negate the adverse effects of readily soluble carbohydrates on the extent and rate of degradation in sheep. The result supports the conclusion of Cerrillo et al. (1999) that high quality roughage is less susceptible to the negative effect of readily soluble carbohydrates than low quality roughage when fed to goats and sheep (Matejousky and Sanson, 1995). Recently, Olson et al. (1999) concluded that the negative effect of readily degradable starch on OM digestion of low quality roughage was not fully overcome by the addition of degradable protein.

For CP, parameters A and B were similar for ARS, T1 and T2, and significantly different from URS. Degradation rate (c) of T1 and T2 is significantly different from ARS. Ammoniation resulted in about 17% increase ($p<0.01$) in protein solubility, which was significantly higher than URS. This result

Table 2. *In situ* dry matter, crude protein and neutral detergent fiber degradation characteristics of URS and ARS based-diets supplemented with rice bran

	URS	ARS	T_1	T_2	SEM	Level of significance
Dry matter						
A	21.9^a	24.0^a	32.45^b	29.0^c	1.2	**
B	43.6^a	50.2^b	34.10^c	47.9^b	2.2	*
A+B	65.5^a	74.2^b	65.55^a	76.9^b	1.9	*
c	0.018^a	0.035	0.038^b	0.035^b	0.003	*
ED (0.02)	42.9^a	54.4^b	53.9^b	58.8	1.8	**
(0.05)	34.2^a	42.2^b	45.4^b	47.6^c	1.5	**
Crude protein						
A	36.0^a	53.6^b	50.8^b	49.8^b	1.2	**
B	28.1	30.80	28.4	33.8	2.2	ns
A+B	64.1^a	84.4^b	79.2^b	82.6^b	1.9	**
c	0.024^a	0.036^b	0.06^c	0.05^c	0.004	*
ED (0.02)	51.0^a	72.4^b	71.5^b	72.8^b	2.0	**
(0.05)	44.9^a	64.7^b	65.2^b	64.7^b	1.5	**
NDF						
A	6.8	4.9	5.2	5.8	1.6	ns
B	58.0^a	67.6^b	51.3^a	66.4^b	2.1	*
A+B	64.8^a	72.5^b	56.5^a	72.2^b	2.0	*
c	0.017^a	0.033^b	0.022^a	0.03^b	0.002	**
ED (0.02)	32.8^a	44.5^b	29.6^a	43.6^b	2.0	**
(0.05)	21.0^a	28.2^b	18.7^a	27.7^b	1.3	*

A is the water-soluble components and *B* the insoluble but degradable fractions at *t*; $B=(a+b)-A$.

a, *b* and *c* are constant in the exponential $p=a+b(1-e^{-ct})$.

ED=Effective degradability.

Means with common superscripts within rows are not significant.

Significance level * $p<0.05$); ** $p<0.01$.

URS=Untreated rice straw; ARS=Ammonia treated rice straw.

T_1=65% URS+30% RB+5% SBM. T_2=85% ARS+15% RB.

seems to indicate that ammoniation alone could provide the same effect with that of RB supplementation on protein solubility of straw based-diets. The higher CP solubility (A) is due mainly to ammoniation and RB supplementation that could in turn increase concentration of NH_3-N in the rumen.

Both ammonia treatment of straw and RB supplementation did not affect the insoluble but degradable CP fraction (B) contrary to its effects on DM and NDF. Effective CP degradability also showed insignificant variation among these straw diets. Lines and Weiss (1996) had similar results on incubating ammoniated hay. Ammoniation nor supplementation did not affect degradation of insoluble CP, and that the apparent lag noted between treated and untreated roughage reflect the increase microbial contamination (figure 1). However, with more soluble protein, the potential degradability (A+B) was improved by ammoniation (ARS and T2) and RB supplementation of URS based-diet. As expected, the effects of ammoniation on fiber degradation is more pronounced than the effect of supplementation except for parameter NDF-A where it is not significant. Both the extent and rate of NDF degradability were significantly increased by the treatment. The improvement could be attributed to changes in cell wall structure, particularly from solubility of hemicellulose (Klopfenstein, 1978; Ibrahim et al., 1989). In addition, ammoniation increased microbial accessibility to bonds linking the macromolecules, thereby allowing greater penetration of cellulase enzymes which changes the physical rather than the chemical structure of the cell wall (Innocenti et al., 1989; Mason et al., 1990; Oliveros et al., 1993). These effects reduced the proportion of cell wall polysaccharides recovered by neutral detergent (Drydren and Leng, 1988). Ammonia could have also effected higher degradability of the residual cell wall by intracrystalline swelling of cellulose microfibrils (Tarkow and Feist, 1969; Millet et al., 1976). The significant increase in the extent and rate of NDF degradation in straw could provide more digestible fiber pool for the animals (Oliveros et al., 1993).

Rice bran supplementation did not affect fiber (NDF) degradability of either URS or ARS. This result is consistent with the earlier report of Zhao et al. (1996b), where they observed that RB inclusion reduced ruminal digestibility of mixed by-products diets when fed to steer, but not on its basal components particularly hay or sugar beet pulp which gave similar *in situ* rumen disappearance. Using barley as source of readily fermentable carbohydrate, Huntanen (1988) also found no marked differences in the degradability of alkali-treated straw or grass silage with or with out rolled barley. There could be negative effect of free fatty acids from RB on fiber degradability of cereal straw (Devendra and Lewis, 1974; Farrell, 1994) but this was not observed in this study. Plausibly, the rate of RB supplementation is low to have negative effects of free fatty acids on fiber degradability. In reference, Zhao et al. (1996b) confirmed the conclusion of that the detrimental effect of full fat RB can be observed beyond 30% supplementation.

CONCLUSION

The improvement in the ruminal degradability of rice straw brought about by ammonia treatment was manifested through enhanced accessibility of cell wall components to microbial breakdown. Moreover, it provided more degradable DM and protein in the rumen as a result of the treatment. The improvement in the quality of rice straw through ammoniation reduced the negative effect of rice bran supplementation on DM degradation. Supplementation of RB yielded a positive response to the degradability of straw diet particularly for CP. A 15% inclusion of RB to ARS provided readily soluble carbohydrates and improved protein solubility, the same effect of ammonia treatment. Ammoniation had more pronounced effect on degradation characteristics of fiber fraction than RB supplementation. Ammonia treatment and supplementation of RB can improve rumen degradability of its cell wall components, which could lead to better utilization of straw and more digestible fiber for sheep.

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