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Effect of Urea Treatment on Chemical Composition and Digestion of *Cenchrus ciliaris* and *Cynodon dactylon* Hays and *Zea mays* Residues

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Abstract: Buffelgrass (*Cenchrus ciliaris*), bermudagrass (*Cynodon dactylon*) hays, corn stover and corn cobs (*Zea mays*), with a moisture content of 50%, were ammoniated with feed grade urea at 0, 4.5 and 6.0% DM. Ammonia-treated forages were stored in plastic bags during 21 days. Crude Protein (CP), Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), Acid Detergent Lignin (ADL), hemicellulose and cellulose were determined. The rate and extent of dry matter and NDF loss was estimated using the nylon bag technique. Rumen cannulated Pelibuey×Rambouillet sheep were used to incubate the nylon bags. During the trial, sheep were fed *ad libitum* 70% sorghum Sudan hay and 30% alfalfa hay. The CP content in buffelgrass hay (4.4, 9.9 and 16.3%, respectively) bermudagrass hay (6.2, 13.8, 22.9%), corn stover (7.4, 12.3, 15.6%) and corn cobs (3.0, 11.2, 16.7%) increased as urea treatment augmented. However, NDF was reduced as treatment increased (80.6, 74.7, 75.8; 79.9, 77.8, 75.5; 75.4, 66.0, 69.3; 92.5, 85.6 and 88.4%, respectively). Similar, pattern as NDF was found in hemicellulose content. Effective Degradability of Dry Matter (EDDM) and NDF (EDNDF) in buffelgrass hay (26, 42, 31; 20, 33 and 24%, respectively) and corn cobs 23, 36, 32; 25, 34, 31%, respectively) increased quadratically ($p<0.01$) as urea treatment augmented. Whereas, EDDM and EDNDF in bermudagrass hay (30, 38, 39; 25, 32, 33%, respectively) and corn stover (42, 48, 53; 38, 41, 45%, respectively) increased linearly ($p<0.01$). Urea ammoniation altered the chemical composition and digestibility of forages improving their nutritive value.

Key words: *Cenchrus ciliaris* hay, *Cynodon dactylon* hay, corn stover, corn cobs, urea ammoniation, chemical composition, effective degradability

INTRODUCTION

Buffelgrass (*Cenchrus ciliaris*), bermudagrass (*Cynodon dactylon*) hays, corn stover and corn cobs (*Zea mays*) are produced in northeastern Mexico and are important feed resources for ruminants during dry seasons (Ramirez, 2003). However, the main constrains for their animal use are low voluntary intake, digestibility, nitrogen and energy content and high cell wall concentration.

Feed-grade urea is widely available and has been used as a source of ammoniation to improve the feeding value of various grass hays and crop residues by increasing non protein nitrogen content, fiber digestibility and is an effective preservative or fungicide (Sundstol and Coxworth, 1984). The effect of urea treatment on the nutritive value of roughages is the result of two processes which occur within the treated forage: Ureolysis that turns urea into ammonia through an enzymatic reaction that requires the presence of water and urease enzyme and the effect of ammonia on the cell walls of forage. Several factors such as urea doses, moisture and temperature affect the effectiveness of urea treatment. The enzyme urease is produced by bacteria naturally occurring on plant material, but water must be present for urease to break down urea into two ammonia ions (Brown and Adeji, 1995).

When limited amounts of hay or other roughages are available, ammoniation may be a cost-effective way to increase the value of forages. However, ammoniated feeds should be analyzed prior to feeding to determine actual nutrient content. Energy supplementation may still be necessary after ammoniation, depending on the nutrient requirements of each particular set of livestock. Ammoniated straw has a feeding value similar to average-quality grass hay (Landy and Anderson, 2005).

The aim of this study, was to determine the chemical composition and the degree and extent fiber digestion of four forages untreated and treated with different urea levels.

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MATERIALS AND METHODS

Buffelgrass and bermudagrass hays and corn stover and corn cobs were obtained from a commercial store at Monterrey City at the State of Nuevo Leon, Mexico. Samples of roughages were chopped (approximately 1 cm length). Feed grade urea dissolved in water equivalent to 0, 4.5 and 6.5% (w/v) were sprayed on 100 g quadruplicate samples on Dry Matter (DM) basis. Water was added to increase the moisture content to 50%. The samples were thoroughly mixed rapidly and stored in plastic containers, sealed and kept for 21 days at ambient temperature (24-26°C). At the end of storage period, samples were exposed for 2 days for excess NH_3 escape. They were ground through 1 mm screen with a hammer mill and crude protein (AOAC, 1997), Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), Acid Detergent Lignin (ADL) were determined (Van *et al*., 1991). Hemicellulose (NDF-ADF) and cellulose (ADF-ADL) were calculated by difference.

The rate and extent of DM and NDF loss was estimated using the nylon bag technique. Four rumen cannulated Pelibuey×Rambouillet sheep (weighing 45±3.5 kg, BW) were used to incubate the nylon bags (5×10 cm, 53 μm of pore size), which contained ground (4 g) samples of each roughage. During the trial, sheep were fed 70 % sorghum Sudan hay and 30% alfalfa hay *ad libitum*. Incubation periods were at 4, 8, 12, 24, 48, 72 and 96 h. Upon removal from the rumen, bags were washed in cold water. Zero time disappearance was obtained by washing unincubated bags in similar fashion and then bags were dried at 55°C in an oven for 48 h. Disappearance of DM and NDF for each incubation period was calculated by:

$$\text{Disappearance, \%} = \frac{\text{(Initial weight - final weight)}}{\text{(initial weight)}} \times 100$$

Digestion characteristics of DM and NDF which were estimated by equation of Orskov and McDonald (1979):

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

Where:

p - Is disappearance rate of DM and NDF at time t,
a - Is an intercept representing the portion of DM and NDF solubilized at the beginning of incubation (time 0),
b - Is the portion of DM and NDF that is slowly degraded in the rumen,
c is the rate constant of disappearance of fraction b
t - Is the time of incubation.

The nonlinear parameters a, b and c and effective degradability of DM (EDDM) and NDF (EDNDF) = $(a+b)c/(c+k)(e^{-(ct)LT})$, were calculated using the Neway computer program (McDonald, 1981); k is the estimated rate of rumen out flow and LT is the time lag. The EDDM and EDNDF of samples were estimated assuming a rumen out flow rate of 5 % h^{-1}.

Parameters of *in situ* digestibility: a, b, c and EDDM and EDNDF of each kind of roughage were statistically analyzed using a Latin Square design 4×4. Means were compared using orthogonal polynomials ($p<0.05$), linear and quadratic effects were reported (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

The Crude Protein (CP) content in buffelgrass hay, bermudagrass hay, corn stover and corn cobs was increased as urea treatment augmented from 0 to 6.5% (Table 1). Increments in CP concentration may be due to non-protein-nitrogen addition to the plant particles.

Table 1: Chemical composition of the buffelgrass and bermudagrass hays, corn stover and corn cob treated with different urea levels

	Urea treatment level		
Concept	0%	4.5%	6.5%
	Buffelgrass hay		
Crude protein, %	4.4	9.9	16.3
Neutral detergent fiber, %	80.6	74.7	75.8
Acid detergent fiber, %	51.7	56.3	54.6
Cellulose, %	38.1	42.5	39.6
Hemicellulose, %	28.9	18.4	21.2
Acid detergent lignin, %	10.0	10.5	12.3
Insoluble ashes, %	3.6	3.3	2.7
Cell content, %	19.4	25.3	24.2
	Bermudagrass hay		
Crude protein, %	6.2	13.8	22.9
Neutral detergent fiber, %	79.9	77.8	75.5
Acid detergent fiber, %	41.1	43.8	44.4
Cellulose, %	30.3	32.7	33.6
Hemicellulose, %	38.8	34.0	32.1
Acid detergent lignin, %	7.3	7.8	7.3
Insoluble ashes, %	3.5	3.3	3.5
Cell content, %	20.1	22.2	24.5
	Corn stover		
Crude protein, %	7.4	12.3	15.6
Neutral detergent fiber, %	75.4	66.0	69.3
Acid detergent fiber, %	46.7	43.0	44.0
Cellulose, %	36.8	34.6	36.5
Hemicellulose, %	28.7	23.0	25.3
Acid detergent lignin, %	8.9	7.3	6.1
Insoluble ashes, %	1.0	1.1	1.4
Cell content, %	24.6	34.0	30.7
	Corn cobs		
Crude protein, %	3.0	11.2	16.7
Neutral detergent fiber, %	92.5	85.6	88.4
Acid detergent fiber, %	49.4	54.1	54.2
Cellulose, %	37.3	42.2	42.2
Hemicellulose, %	43.1	31.5	34.2
Acid detergent lignin, %	11.5	11.5	11.4
Insoluble ashes, %	0.6	0.4	0.6
Cell content, %	7.5	14.4	11.6

Table 2: Means of *In situ* digestibility parameters and effective degradability of dry matter of buffelgrass and bermudagrass hays, corn stover and corn cob treated with different urea levels

	Urea treatment level				Sig	
Concept	0%	4.5%	6.5%	SEM	L	Q
	Buffelgrass hay					
a, %	9.2	14.1	12.1	0.2	ns	**
b, %	36.3	56.9	44.0	5.0	ns	ns
c, % h^{-1}	4.0	6.0	3.0	1.0	ns	ns
EDDM, %	26.2	41.6	31.2	0.6	ns	**
	Bermudagrass hay					
a, %	11.1	10.6	13.0	0.2	*	ns
b, %	39.4	55.9	53.1	1.5	ns	*
c, % h^{-1}	5.0	6.0	6.0	0.4	ns	ns
EDDM, %	30.3	38.7	39.2	0.5	**	ns
	Corn stover					
a, %	18.1	24.1	23.4	1.0	ns	ns
b, %	52.3	52.3	53.5	1.6	ns	ns
c, % h^{-1}	4.0	4.0	5.0	0.01	ns	ns
EDDM, %	41.9	48.0	52.8	1.5	*	ns
Corn cobs						
a, %	1.1	5.7	6.6	0.6	*	ns
b, %	63.0	68.4	72.7	2.4	ns	ns
c, % h^{-1}	3.0	4.0	3.0	0.2	ns	ns
EDDM, %	22.9	35.6	32.1	0.9	ns	*

a = fraction of dry matter lost during wash; b= fraction of dry matter degraded; c= rate of degradation of dry matter; EDDM= Effective Degradability of Dry Matter, considering a rumen outflow rate of 5 % h^{-1}; SEM= Standard Error of the Mean; Sig = significant level; L = linear; Q = Quadratic; *($p<0.05$); **($p<0.01$); ns = not significant

Fall (1988) and Aregherore (2005) also reported that corn stover increased their CP content from 6-15% when treated the roughage with 0 and 7% feed grade urea. However, Golmahi *et al*. (2006) found a small increment (4%) in CP of barley straw treated with urea at 0 and 4%.

In this study, as urea treatment increased, NDF and hemicellulose decreased and ADF, ADL and insoluble ashes remained about the same (Table 1). The NDF content of all roughages might have decreased as a result of hemicellulose solubilization (Sundstol and Coxworth, 1984). Previous studies have also reported reduction of NDF in urea-treated crop residues and grass hays. Rodriguez *et al*. (2002) found that *Brachiaria humidicola* hay ammoniated with a urea solution at 6.0%, NDF content decreased compared to untreated hay. Moreover, Cañeque *et al*. (1988) argued that NDF concentration was reduced in barley straw treated with urea at 6.0%. In addition, maize residues reduced their cell wall content when treated with urea at 7% DM (Aegheore, 2005; Oji *et al*., 2007). In this study, *in situ* digestibility parameters of dry matter and EDDM in all roughages are shown in Table 2. The soluble fraction of the DM and EDDM in Buffelgrass hay increased quadratically ($p<0.01$) as urea treatment augmented. The degradable fraction and rate constant of degradation of DM were unaffected ($p>0.05$) by urea treatment. The same pattern was found in corn cobs. Gobbi *et al*. (2005) also reported that *Brachiaria decumbens* hay, treated with urea at 0, 2, 4, 6, 8 and 10% of the forage DM, increased quadratically the *In vitro* Dry Matter Digestibility (IVDMD) reaching the maximum digestibility of 69% at the 7% urea level. In this study, EDDM in bermudagrass hay ($p<0.01$) and in corn stover ($p<0.05$) augmented linearly, as urea treatment increased up to 6.5%. Brown and Adjei (1995) also found *in vitro* DM digestibility increased linearly ($p<0.01$), whereas concentrations of hemicellulose and ADL decreased linearly ($p<0.05$) with increasing urea level in *Panicum maximum* hay treated with urea at 0, 4, 6 or 8% of the forage DM.

Other studies (Dias-De-Silva *et al*., 1988; Chesnot *et al*., 1991; Joy *et al*., 1992) proved that, in general, a dosage of 4-6% urea can be considered sufficient to obtain maximum digestion of dry matter and reduction of NDF, when compared to untreated corn stover and when the treatments are carried out with a moisture content of 30-50%. In addition, Brown and Kunkle (1992) when ammoniated *Cynodon dactylon* hay with 4% feed grade urea reported that NDF decreased and *in vitro* DM digestibility increased.

Moreover, Hill and Leaver (1999) reported that application of urea at a rate 4% (DM) increased ($p<0.01$) reduction in NDF content compared to wheat straw ensiled without urea. And treatment of harvested wheat straw at ensiling with urea reduced ($p<0.001$) the conversion of particular sugars to fermentation acids during conservation but slightly enhanced the loss of starch. They concluded that treatment of whole-crop wheat forage with urea at harvest can be considered an effective method of preserving forage nutritive value. In other study, Granzin and Dryden (2003) examined the

Table 3: Means of *In situ* digestibility parameters and effective degradability of neutral detergent fiber of buffelgrass and bermudagrass hays, corn stover and corn cob treated with different urea levels

Concept	Urea treatment level 0%	4.5%	6.5%	SEM	Sig L	Q
	Buffelgrass hay					
a, %	1.6	3.4	0.2	0.3	ns	*
b, %	40.0	61.2	59.1	4.9	ns	ns
c, % h^{-1}	4.0	6.0	3.0	0.5	ns	ns
EDDM, %	20.3	33.1	23.7	0.9	ns	**
	Bermudagrass hay					
a, %	2.0	0.6	1.2	0.7	ns	ns
b, %	45.0	61.6	63.2	1.4	*	ns
c, % h^{-1}	6.0	6.0	6.0	0.2	ns	ns
EDDM, %	24.8	32.0	33.0	0.5	**	*
	Corn stover					
a, %	9.6	8.8	8.6	1.2	ns	ns
b, %	59.7	70.9	71.8	0.7	**	ns
c, % h^{-1}	5.0	5.0	6.0	1.0	ns	ns
EDDM, %	38.3	40.6	45.2	1.8	*	ns
	Corn cobs					
a, %	1.2	0.9	1.7	0.7	ns	ns
b, %	64.7	75.3	78.2	2.2	*	ns
c, % h^{-1}	3.0	4.0	4.0	0.1	ns	ns
EDDM, %	25.0	34.3	31.4	0.8	ns	*

a = fraction of neutral detergent fiber lost during wash; b = Fraction of neutral detergent fiber degraded; c = rate of degradation of neutral detergent fiber; EDNDF = Effective Degradability of Neutral Detergent Fiber, considering a rumen outflow rate of 5 % h^{-1}; SEM= Standard Error of the Mean; Sig = significant level; L = Linear; Q = Quadratic; *(p<0.05); **(p<0.01); ns = not significant

effects of urea (0, 2, 4, 6 and 8% DM) and water (250, 500 and 750 mL kg^{-1} DM) treatment on rhodes grass (*Chloris gayana* cv. Callide) hay. The combinations of water and urea that resulted in the highest concentrations of CP (28%) and organic matter digestibility (75%) were 500 mL of water and 8% urea. They concluded that urea significantly alters the NDF content and digestibility of rhodes grass hay and urea also increases its CP content. In addition, Abebea *et al*. (2004) found that NDF decreased and *in vitro* DM digestibility increased when ammoniated wheat straw with a solution of feed grade urea at 5.4% of straw DM.

Previous research studies have reported positive performance in animals feeding urea-treated crop residues. In a study, Sharma *et al*. (2004) carried out with 48 multiparous buffaloes, maintained under a subsistence-oriented mixed farming system, to assess the effect of feeding 4% urea-treated wheat/rice straw with a supplement (parts/100:deoiled soybean cake 10 to12; rice polish 88-90) during late pregnancy (about 8-10 weeks before calving) and lactation (300 d), animals significantly improved straw consumption, milk yield, body condition and the level of cash benefits. Moreover, Verma *et al*. (2006) reported that feeding of with 4% urea ammoniated wheat straw showed significantly higher DM intake, digestibility and balances of nutrients in crossbred cattle, compared to the untreated wheat straw fed group. They recommended the use of ammoniated wheat straw in the diets of cattle to economize the cost of concentrate by reducing 22% deoiled groundnut cake. In addition, Oji *et al*. (2001) investigated changes in chemical composition and nutritive value of corn residues untreated and treated with urea at 3.0%. They found that treatment improved (p<0.05) digestion coefficients of CP, DM, NDF, ADF and organic matter in goats and recommended that feed grade urea can be used to improve the nutritional value of corn residues for small ruminants feeding during off season periods.

In situ digestibility characteristics of NDF and EDNDF (Table 3) followed the same pattern as those showed in the DM content of all roughages. Higher EDNDF in buffelgrass hay and corn cobs was obtained at 4.5% urea treatment; conversely, the NDF in bermudagrass hay and corn stover was digested in greater extent at 6.5% urea treatment. Guedes *et al*. (2000) also found that application of urea to whole-crop triticale at rates of 60 and 90 g kg^{-1} DM significantly (p<0.05) reduced the NDF content and increased (p<0.05) the NDF *in vitro* digestibility when compared to untreated materials. Whole-crop triticale harvested at later growth stages (approximately 60% DM) can be effectively preserved and upgraded by ensiling with 60-90 g urea kg^{-1} DM.

It has been found that the more impacting changes on the nutritional value of urea-treated roughages are observed in those plant materials with high NDF (Rodriguez *et al*., 2002; Guedes *et al*., 2006), In this study, buffelgrass hay and corn cobs, with higher NDF content, resulted with higher increments in CP (Table 1), EDDM (Table 2) and EDNDF (Table 3) than bermudagrass hay and corn stover. The reasons adduced for improvements after ammoniation of different roughages evaluated here and elsewhere, include collapse of vascular bundle sheath cells, rupture of inner cuticular surfaces and separation from adjacent ground parenchyma, alteration in the friability of the rigid layer converting the inner surface of cell wall and the ability that NH_3 has to dissolve parts of hemicellulose cleaving ester bonds of uronic acids with loss of acetyl groups, thus releasing acetic and phenolic acids, an effect that may possible explain the decreases in NDF (Rodriguez *et al.*, 2002; Brown, 1993).

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CONCLUSION

Ammoniation improved the feeding value of the all evaluated roughages by increasing the N content and promoted the chemical breakdown of plant fibers resulting in a better opportunity, for rumen microbes to attach to the fiber and digest the ammoniated plant material in the rumen of sheep. Neutral detergent fiber concentration, which is a measure of the cell wall content of forage, was reduced by ammoniation. This contributed to the greater DM digestion of ammoniated compared to untreated roughages. However, the more impacting changes on the nutritional value of urea-treated roughages were observed in plant materials of less quality such as buffelgrass hay and corn cobs which resulted with higher increments in CP, EDDM and EDNDF compared to bermudagrass hay or corn stover that had less NDF content. Dry matter digestion is a measure of the energy content of forage. Thus, results obtained here and elsewhere suggest the use of urea at 4.5 to 6.5% with a moisture content of 50%, for upgrading the N and energy availability of poor quality grass hays and corn residues. However, its proportion in the diet of the ruminant should be limited so as to avoid risk of ammonia toxicity or transmission of toxic compounds into milk. More research work is needed to evaluate accurately the daily optimal consumption of urea-treated forages produced in NE Mexico.

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