In Vivo Digestibility of cellulase-treated rice Straw in Water Buffalo (*Bubalus bubalis* Linn.)

N.R. Bariroh

Assessment Institute of Agricultural Technology of East Kalimantan
Jl. Pangeran M. Noor, Sempaja – Samarinda, East Kalimantan
rizqibarir@yahoo.com

Fibrous crop represent by far the majority of total volume of feeds produced in Southeast Asia and constitute the basic feed sources for most crop animal system. These fibrous crop residues are poor sources of fermentable nitrogen and their crude protein is below the level required by rumen microorganism. Besides, they are low in easily degradable carbohydrates and minerals. All of these result in limited intake, poor rumen function and increased methane emissions to the atmosphere. This study was conducted to determine the in vivo digestibility of nutrients in cellulase-treated rice straw, at the Philippines Carabao Center Farm, University of the Philippines Los Banos, College at Laguna from april - may 2004. Research design used was Randomized Complete Block Design with 3 treatments and 3 replications. Nine growing buffalos were fed a diet of 60% rice straw 40 % brewer's spent grain. Rice straw was subjected three treatments : treatment 1 (0% cellulase), treatment 2 (0.25% cellulase), and treatment 3 (0.5% cellulase). The result show that cellulose increased the apparent DM digestibility by 6.86 % and 4.88%, crude protein by 3.16 % and 4.9%, crude fiber by 8.63% and 9.14%, in treatment 2 and 3 respectively.

Key words : *buffalo, cellulase, rice straw, in vivo digestibility*

Tanaman serat merupakan pakan terbesar yang diproduksi di Asia Tenggara dan merupakan merupakan sumber pakan dasar pada sebagian besar sistem integrasi ternak tanaman. Limbah tanaman serat ini rendah nitrogen yang dapat difermentasi dan protein kasarnya di bawah level yang dibutuhkan mikroorganisme rumen. Di samping itu limbah pangan mempunyai karbohidrat yang dapat dicerna dalam dosis rendah, karena adanya asupan yang rendah dan fungsi rumen yang rendah, pada akhirnya meningkatkan emisi methan ke atmosfer (Studi ini dilaksanakan di UPLB Laguna dari april-mei 2004). Rancangan percobaan yang digunakan adalah RAK dengan 3 perlakuan dan 3 ulangan. Sembilan kerbau dalam masa pertumbuhan diberi pakan yang terdiri dari jerami padi sebesar 60%, 40% limbah pabrik bir. Jerami padi diperlakukan : perlakuan 1 (0% selulase), perlakuan 2 (0. 25 selulase dan perlakuan 3 (0,5% selulase). Hasil studi membuktikan bahwa selulase meningkatkan kecernaan bahan kering sebesar 6.86 % dan 4.88%, protein kasar sebesar 3.16 % dan 4.9%, serat kasar sebesar 8.63% dan 9.14%, masing masing pada perlakuan 2 dan 3.

Introduction

Fibrous crop residues play an important role in Southeast Asia. These are poor sources of fermentable nitrogen, and their crude protein is below the level required by rumen microorganism (Cheeke, 1999). Besides they are low in easily degradable carbohydrates and minerals. All of these result in limited intake, poor rumen function and increased methane emission to the atmosphere (Pezo and Devendra, 2001).

Rice straw, one of fibrous crop residues, is potential source of feed because straw consists of structural (cell wall) polysaccharides such cellulose, hemicelluloses and pectin, which are the major components of the ruminant diet and are the primary source of energy in forage based diets (Wang and Mc Allister, 2002). The problem with using rice straw, however, is its low digestibility. Without treatment, the digestibility of rice straw is so low that it has little value as feed (Cheeke, 1999). This can be improved by grinding or chopping the material to reduce particle size, exposing more terminal ends of cellulose fibers to cellulase. One potential strategy to increase the digestibility of rice straw, especially cellulose is by using fibrolytic enzyme. Feng et al (1996) noted that the application of enzyme immediately before feeding increased digestibility of feed. This study aimed to determine the in vivo digestibility of nutrients on diet containing cellulose treated rice straw by water buffalo.

Methode

Time and place of study

This study was conducted at the Philippines Carabao Center Farm, University of the Philippines Los Banos, Laguna, from April to May 2004.

Animal

Nine growing water buffalos (1-2 years old and 205-262 kg of live weight) were used. All buffaloes were placed in individual stall during experiment. Dewormer was given in the beginning of the experiment. The animals were provided with a mineral block as well as water.

Diet

The diet consisted of 60% of rice straw and 40% brewer's spent grain. The rice straw was treated with cellulase, with the untreated diet regarded as the control. The nutrient levels were in accordance with the feeding standard table as recommended by Kearl (1982). The application of cellulase onto rice straw was by chopping rice straw into 5 cm pieces and spraying with cellulase solution, prior to feeding. This ration was offered twice daily at 8.00 am and 3.00 pm. Brewer's spent grain was placed in a separate container to facilitate the proper collection and sampling of feed refusal. An adjustment period of seven days was given to accustom the animals to the cages of stalls. The 14 succeeding days were the preliminary period. During this period the animals were given test ration in order to eliminate the problem of selective eating, which could lead to refused feed (orts). Another purpose of the preliminary period was for the digestive tracts of the animals to be thoroughly adjusted to the ration being tested. After the preliminary period, samples were collected for five days.

Source of enzyme

The enzyme was produced by the National Institute of Molecular Biology and Biotechnology, Los Banos. The enzyme activity of cellulase was 4800 IU / ml.

Treatment and procedure

Three treatment were tested, namely :

Treatment 1 = 60% of rice straw + 40% brewer's spent grain

Treatment II = 60 % rice straw treated with 0.25% cellulase (2.5 ml/kg DM) + 40% brewer's spent grain

Treatment III = 60 % rice straw treated with 0.5% cellulase (5 ml/kg DM) + 40% brewer's spent grain

Feed refusals (orts) and feces were collected daily at 7.30 a.m. and weighed. At the end of each collection time, the feeds, orts and fecal samples were dried immediately using a forage drier. At the end of each collection period, the dried samples of feeds, orts and feces were composited and ground, and appropriate

representative amounts were taken for laboratory analysis. All samples of feed offered, orts and feces were analyzed for dry matter, crude protein, crude fiber (AOAC, 1993).

Digestibility

Apparent digestibility was determined using formula below .

$$\% \text{ digestibility} = \frac{\text{amount of nutrient consumed} - \text{amount of nutrient excreted in feces}}{\text{Amount of nutrient consumed}}$$

Statistical analysis

The statistical design used was the Randomized Complete Block Design (RCBD). The result were analyzed using MSTAT software (1990).

Result and Discussion

Nutrient composition of the feed offered attached in Table 1. Nutrient content of rice straw in this study were different to the result reported by Gerpacio and Castillo (1979) who found that the nutrient content of rice straw was 92.6% dry matter, 31.14% crude fiber, 1.44% crude fat, 21.21 % ash. The protein content of rice straw used in this study was not similar with Juliano et al. (1988) that reported that protein content of rice straw was 4.9%. The difference of nutrient content could be attributed to factors like species/variety, soil, environment, and management/cultural practices (Roxas, 1988). Furthermore, Gerpacio and Castillo stated that nutritive value of brewer's spent grain was 93.5% dry matter, 21,8% crude protein, 4.9% crude (PCARRD, 1994).

Table 1. Nutrient composition of the feeds (% dry basis)

Component	Rice straw	Brewer's spent grain
Dry matter (as fed)	73.54	21.49
Ash	25.39	4.6
Crude Protein	6.95	36.06
Crude fiber	30.06	13.8
Crude fat	1.54	9.92

It can be seen from Table 2. that the digestibility of cellulase-treated rice straw was higher than that of untreated rice straw, with cellulase increasing dry matter digestibility by 6.86 % and 4.88%. Feng at al (1996) reported that in vivo dry matter digestibility was higher when exogenous fibrolytic enzymes were applied to forage than in untreated forage.

Table 2. Effect of cellulase on apparent digestibility of dry matter, crude protein, crude fiber

Component	Cellulase in Rice Straw, %		
	0	0.25	0.5
Dry matter	43.67 [b]	50.53 [a]	48.55 [a]
Crude protein	65.31 [b]	69.47 [a]	70.23 [a]
Crude fiber	39.00 [b]	47.63 [a]	48.14 [a]

[a, b] Row means with different superscripts were significantly differ (P<0.05)

Cellulase

Straw which is treated with cellulase

Stokes and Zheng (1995) reported that spraying enzymes on forage increased dry matter intake by 10.7% and milk yield by 14.7%. One advantage of such a method is that it may partially protect fibrolytic enzyme from ruminal degradation when they are sprayed onto feeds, because binding with substrates may cause conformational changes that may protect these exogenous enzyme from ruminal protease. Morgavi et al (2000) suggested that synergy between ruminal fibrolytic enzyme (with pH optimum below 6.0) may be responsible for improvement in animal production when ruminant are fed feeds treated with enzymes.

Furthermore, Joblin et al. (1989) reported that rumen fungi were able to gain access to plant polysaccharides that were unavailable to rumen bacteria. It is possible that at high levels of cellulase preparation, some of the many structural barriers to digestion of feed were removed by enzymatic action, allowing the bacteria to colonize plant fiber and out of compete the fungi.

Table 2. shows that enzyme-treated rice straw exhibited significantly higher apparent protein digestibility by 3.16% and 4.92% for 0.25% and 0.5% of cellulose, respectively. Yang et al. (1999) reported that a change in ruminal protein metabolism with the use of fibrolytic enzymes. Ruminal degradability of feed protein increases, with a concomitant increase in microbial protein synthesis. It also increased crude fiber digestibility by 8.63% and 9.14% at cellulose levels of 0.25 % and 0.5%, respectively. The reduced crude fiber content in cellulase-treated straw compared to the untreated control was a result of enzymatic solubilization of plant fiber (Hristov et al., 1998).

Performance of Philippines

Financial analysis

Financial analysis of treated and untreated rice straw attached in Table 3.

Table 3. Financial analysis of treated and untreated rice straw effect to buffalo (for 1 animal) for a month

Component	Untreated rice straw (Rp)	Treated rice straw (Rp)
Variable cost		
- Feed	150,000	150,000
- Enzyme		13,000
- Labor	100.000	100,000
Total	250,000	
Fixed Cost		
- Stall reduction	4,167	4,167
- Equipment reduction	9,375	9,375
Total		
Revenue	330000	349.500
R/C	1.25	1.26

Assumption : - Daily weight gain of buffalo fed with treated rice straw feed 6% higher (daily weight gain 0.4 kg/day)
- The price of live weight Rp 27.500/kg

Conclusion

Cellulase increased the apparent DM digestibility by 6.86 % and 4.88%, organic matter by 5.36% and 4.15%, crude protein by 3.16 % and 4.9%, crude fiber by 8.63% and 9.14%, in treatment 2 (2.5 ml/kg DM of rice straw) and 3 (5 ml/kg DM of rice straw) respectively.

Literature Cited

Cheeke, P.R. 1999. Applied animal nutrition. Feeds and Feeding. Second edition. Prentice hall, Inc. New Jersey.

Feng, P., C.W. Hunt, G.T. Pritchard and W.E. Julien. 1996. Effect of enzyme preparation on in situ and in vitro digestive characteristics of mature cool-season grass forage in beef steers. J. Anim. Sci. 74:1349-1357

Hristov, A.N., T.A. Mc Allister and K. Cheng. 1998. Effect of dietary or abomasal supplementation of exogenous polyssacharide-degrading enzymes on rumen fermentation and nutrient digestibility. J. Anim. Sci. 76:3146-3156

Joblin, K.N, A.J. Campbell, A.J. Richardson and C.S. Stewart. 1989. Fermentation of barley straw by anaerobic rumen bacteria and fungi in an anexic culture and in co-culture with methanogens. Lert. Apll. Microbiol. 9:159-197

Kearl, L.C. 1982. Nutrient requirement of ruminant in developing countries. Logan. Utah:International Feedstuffs Institute. Utah Agricultural experiment Station. Utah State University

Morgavi, D.P., K. A. Beauchemin, V.L. Nsereko., L.M. Rode. A.D. Iwaasa, W.Z. Yang, T.A. Mc Allister and Y.Wang. 2000. Synergy between ruminal fibrolytic enzymes and enzymes from *trichoderma longibrachiatum*. J. Dairy Sci. 83:1310-1321

PCARRD. 1994. The Philippines recommends for beef cattle production. Philippine Agriculture and Resources Research Foundation, Inc. Los Banos, Laguna.

Pezo, D.A. and Devendra. 2001. Characteristics of the crop-animal systems practiced in Southeast Asia. International Livestock Research Institute.

Roxas, D.B., M.G. Aller and B.O. Juliano. 1988. Changes in chemical composition and in vitro digestibility of straw components from four rice straw. *In* Ruminant Feeding systems utilizing fibrous agricultural residues – 1987. International Development Program of Australian Universities and College Limited. Canberra.

Stokes, M.R and S. Zheng. 1995. The use of carbohydrase enzyme as feed additives for early lactating cows. In 23rd Biennial Conference on Rumen Function. Chicago, IL. P35 (abstr)

Wang, Y., and T.A. Mc Allister. 2002. Rumen microbes, enzymes and feed digestion-a review. Asian-Austr.J.Anim, Sci. 15:1659-1676

Yang, W.Z., Beauchemin and L.M. Rode. 1999. Effects of an enzyme feed additives on extent of digestion and milk production of lactating dairy cows. J. dairy Sci. 82 : 391-403