Asia-Pacific Journal of Science and Technology: Volume: 27. Issue: 06. Article ID.: APST-27-06-07. Research Article

APST

Asia-Pacific Journal of Science and Technology

https://www.tci-thaijo.org/index.php/APST/index

Published by the Research and Technology Transfer Affairs Division,
Khon Kaen University, Thailand

Influence of various treatment methods to improve the quality and utilization of treated rice straw in swamp buffaloes

Pichad Khejornsart[1,*] and Theerayut Jantanam[1]

[1]Faculty of Natural Resources and Agro-Industry, Kasetsart University, Chalermphrakiat Sakon Nakhon Provinces Campus, Sakon Nakhon, Thailand
[*]Corresponding author: fnapck@ku.ac.th

Received 11 June 2021
Revised 12 July 2021
Accepted 2 August 2021

Abstract

In this study, the effect of treated rice straw on nutrient intake, rumen fermentation parameters, and population of microorganisms in the rumen of swamp buffaloes (*Bubalus bubalis*) was investigated. According to the 4 × 4 Latin square design, four yearling swamp buffaloes were randomly assigned to receive a concentrate at 0.5% BW and were offered *ad libitum* with roughage, untreated rice straw (RS), rice straw treated with urea (UTS), rice straw treated with yeast (YTS), and rice straw treated with urea-lime (ULTS). Dry matter feed intake and NH_3-N concentration in the rumen were found to considerably differ among dietary treatments, and 14.0% increased digestibility was observed for the swamp buffalo fed with YTS (p<0.05). Rumen total VFAs and C3 concentration of swamp buffaloes were significantly different after being fed with treated rice straw. However, rice straw treatment did not exert any effect (p>0.05) on the molar proportions of C2 and C4 in swamp buffaloes. The population of predominant cellulolytic bacteria was increased after being fed with YTS or ULTS. In conclusion, swamp buffaloes fed with yeast-treated rice straw could well improve rumen fermentation efficiency and cellulolytic bacteria populations in the rumen.

Keywords: Swamp buffalo, Rumen microorganism, Treated rice straw, Yeast, Molecular technique

1. Introduction

Livestock production, particularly buffalo production, is integrated into the socioeconomic fabric of smallholder farmers and sustainable agricultural systems, especially among the landless rural poor. They have widespread applications, including power, high-protein foods such meat and milk, and organic fertilizers [1-4]. Despite the limitation of tropical feeding resources in terms of quality and quantity, particularly during the dry season, their contribution is essential. In particular, buffaloes and cattle are kept as part of crop production, and rice has been the most widely available crop, with by-products being utilized as feed [5-7]. In rice-producing Asian countries such as Bangladesh, China, India, Indonesia, Vietnam, and Thailand, rice straw is the major agricultural residue. Rice straw exhibits a low crude protein content and a high lignin content, leading to reduced nutrient intake and digestion rates [8]. For several years, researchers have been investigating various physical, chemical, and microbiological methods to improve rice straw quality. Rice straw can be fed to ruminants by various physical and chemical methods [9,10]. Quality improvement by biological treatment compared to chemical approaches is a fascinating solution as it is less harmful to the atmosphere and safer for animals. Wang et al. [11] have reported the use of yeast inoculants to improve cereal straw consumption and ruminant efficiency. Rumen bacteria can convert a significant proportion of structural carbohydrates in fibrous crop residues into volatile fatty acids (VFAs), which is a considerable disadvantage when using this resource as feed. As a result, to adjust for the disadvantages and enhance the feeding efficiency, appropriate treatments and/or supplements are required [12]. Previously, swamp buffalo has been reported to utilize feed more effectively than cattle when maintained under the same conditions, and the diversity of rumen microorganisms increases when swamp buffaloes are fed different types of roughage [13,14]. Hence, new molecular methods with the accuracy and enumeration of rumen microorganisms have been typically employed. However, there are a relatively limited number of studies on rumen fermentation efficiency and microorganisms that are affected by rice straw treatments using molecular approaches. Therefore, the objective of this study is to determine the effect of different types of treated rice straw on the feed intake and rumen fermentation in swamp buffaloes, as well as on rumen microbe composition, by using real-time PCR.

2. Materials and methods

2.1 Preparation of treated rice straw

This research was conducted from March 2020 to June 2020. A combination harvester was used to retrieve the rice straw, which was then thrashed to remove the grain at the maturity stage. The treated rice straw containing urea or a urea-lime mixture was prepared according to that reported by Wanapat et al. [15], involving the mixing of 2 kg each of urea and lime with 100 L of water, pouring it over 100 kg of rice straw, and covering it tightly using a plastic sheet for at least 21 days. Next, 5 L of a *Saccharomyces cerevisiae* starter (1.0×10^8 CFU/mL) was diluted with 3 kg of molasses and 1 kg of urea in 100 L of yeast-treated rice straw, which was then applied to 100 kg of rice straw and fermented for 21 days.

2.2 Experimental design and management of animals

This study was conducted in the Animal Science Research Farm (Kasetsart University, Chalermphrakiat Sakon Nakhon Province Campus, Thailand). Experiments were conducted in compliance with the accurate procedures stated in the Kasetsart University Guide for the Care and Use of Animals (No. ACUC-KU 004/2019). Four yearling swamp buffaloes (*Bubalus bubalis*) (269 ± 28 kg of BW) were randomly assigned to a 4 × 4 Latin square design over four 21-day period. During the experiment, the buffaloes were housed in separate pens with free access to water and mineral blocks. The four rice straw treatments were as follows: I untreated (RS), (ii) 2% urea (UTS), (iii) yeast (YTS), and (iv) 2% urea + 2% lime (ULTS). Concentrate was provided twice a day at a BW of 0.5% at 07:00 and 16:00, with ad libitum access to treated or untreated straw. The concentrates comprised 14% CP and 65% TDN with required levels of minerals (Table 1).

Table 1 The concentrate mixture and the nutritive value (%) as well as treated rice straw.

Items	Concentrate	RS	UTS	YTS	ULTS
Ingredients					
Cassava chip	60.0	-	-	-	-
Rice bran	4.5	-	-	-	-
Brewer's grain	4.0	-	-	-	-
Palm kernel meal	12.0	-	-	-	-
Soybean meal	8.0	-	-	-	-
Leucaena leaf meal	8.0				
Molasses	2.0	-	-	-	-
Urea	0.5	-	-	-	-
Mixed minerals	0.5	-	-	-	-
Salt	0.5	-	-	-	-
Chemical composition, %DM					
DM	89.6	90.6	55.2	50.9	54.9
OM	91.1	87.2	89.8	88.9	87.4
CP	14.2	2.3	5.8	6.2	5.6
NDF	15.8	78.5	70.4	68.7	69.9
ADF	9.5	60.7	56.9	51.5	52.5

RS = untreated, UTS = 2% urea, YTS = yeast, ULTS = 2% urea + 2% lime, DM = dry matter, OM = organic matter, CP = crude protein, EE = ether extract, NDF = neutral detergent fiber, ADF = acid detergent fiber

2.3 Collection of samples and chemical analysis

Feed and refusal were collected for intake evaluation on the last day of each experimental period. Rectal sampling was employed to collect fecal samples. Composites of dietary samples were analyzed for DM, EE, ash and CP content [16], NDF and ADF [17], and acid-insoluble ash (AIA). The AIA content of the feed and refusal was determined by standard procedures [18]. Rumen fluid and content were collected after feeding (0 and 4 h). Rumen pH was immediately measured (HI 8424, HANNA Instruments, USA), and the ammonia nitrogen (NH_3-N) concentration was analyzed by using Kjeltec Auto 1030 Analyzer (Foss Analytical A/S, Denmark). Rumen

VFAs were analyzed by high-pressure liquid chromatography (Agilent 1200 series) using 0.1 M phosphate buffer as the mobile phase. The RBB+C method was employed to distinguish rumen content for DNA extraction [19]. Real-time PCR with specific primers was employed to quantify the populations of the total bacteria, *F. succinogenes*, *R. albus*, and *R. flavefaciens* as described by Koike and Kobayashi [20] (Table 2).

Table 2 Quantitative primers (qPCR) used for the detection of rumen bacteria.

Organisms	Primers	Sequences (5′→3′)	Annealing temperature (°C)	Product size (bp)
Total bacteria	TBF	CGGCAACGAGCGCAACCC	60	141
	TBR	CCATTGTAGCACGTGTGTAGCC		
F.succinogenes	Fs219f	GGTATGGGATGAGCTTGC	63	446
	Fs654r	GCCTGCCCCTGAACTATC		
R. albus	Ra1281f	CCCTAAAAGCAGTCTTAGTTCG	55	175
	Ra1439r	CCTCCTTGCGGTTAGAACA		
R. flavefaciens	Rf154f	TCTGGAAACGGATGGTA	55	295
	Rf425r	CCTTTAAGACAGGAGTTTACAA		

2.4 Statistical analysis

SAS v6.12 was used for statistical analysis (SAS/STAT®; SAS Institute). Analysis of variance was used to analyze the data in a 4 × 4 Latin square design using PROC GLM. Mean values and standard error of the means were utilized to present the results. Difference between treatment means were evaluated using Duncan's New Multiple Range Test [21].

3. Results and discussion

3.1 Chemical compositions of dietary treatment

Table 1 shows the concentrate formulation, three types of treated rice straw, untreated rice straw, and their nutritional values. The nutritional value of rice straw was enhanced by urea, urea-calcium hydroxide, and yeast treatments. Untreated rice straw exhibited a CP of 2.3%, while treated rice straw exhibited CP of 5.6%, 5.8%, and 6.2%, respectively. Remarkably, NDF and ADF proportions were reduced to 69.9%, 68.7%, 52.5%, and 51.5% by urea-calcium hydroxide and yeast treatments. Concentrate components, on the other hand, were derived from locally available resources with a higher CP quality but a low fiber content such as brewer's grain, palm kernel meal and *Leucaena* leaf meal. During the trial periods, the buffaloes had consumed sufficient concentrate. Wanapat et al. [15] have reported that treated and untreated rice straw fed to buffaloes exhibits a percentage composition similar to that of urea-lime treated rice straw. Shen et al. [22] have observed increased fiber degradation as well as a decrease in OM and DM losses in rice straw treated with 5% urea. As reported previously by our group [9,14], the treatment of rice straw with alkali such as sodium hydroxide, urea, and lime leads to the increase in the gas kinetics in *in vitro* trials. Chemical treatment has been reported to improve the digestibility of roughage with a high fiber content, with sodium hydroxide being more effective than urea and calcium hydroxide for degrading the cell walls [15]. The process is essentially delignification, which involves the elimination of linkages between carbohydrates and lignin; hence, a large amount of hemicellulose can be broken down. In addition, structural fibers are physically raised, increasing the number of readable microbial attachment sites on the particle surface, subsequently leading to the increased degradability and feed intake. Urea-treated rice straw treatment is used frequently, which increases the nitrogen content of nonhazardous ensiling materials and serves as a delignifying agent by ammonification, subsequently increasing protein content and nutrients digestibility [14,15,23,24].

3.2 Dry matter intake and nutrient digestibility

Table 3 shows the effect of various treated rice straws on buffalo feed dry matter intake (DMI) and nutritional digestibility. The DMI of rice straw differed significantly among treatments in this study, with the highest DMI observed for yeast-treated rice straw and lowest DMI for untreated rice straw ($p< 0.05$). In case of rice straw treated using yeast and urea-lime, the total DMI in metabolic weight increased to 110.8 and 109.9 g/kg, respectively. Furthermore, results revealed that rice straw treatment significantly improves digestibility. On the other hand, the nutritional digestibility of urea and urea-lime treatments did not differ. Zhang et al. [24] have reported that rice straw pretreated with urea and nitrate leads to increased goat intake and neutral detergent fiber digestibility. Rice straw treated with urea-lime and urea can improve nutrients digestibility, rumen fermentation efficiency, and milk yield, thereby increasing its nutritional value [25]. In addition, Trach et al. [26] have reported that rice straw treated with 5% UTS and 3.5% ULTS leads to increased rumen microbial population and nutritional digestibility. This increase was comparable to the results reported by Wanapat et al. [15]: Feeding ruminants with treated rice straw serves as the optimum nutrition supply for the cellulolytic bacteria of the rumen. However, our group has previously reported [12,14] that chemical improvement by urea, lime or sodium hydroxide did not affect intake of swamp buffaloes. According to the results of this study, the intake of increased dry matter can provide sufficient essential nutrients for the microbial population, such as organic acids, vitamins, and amino acids. Several studies have reported that yeast supplementation improved the digestion of cattle feed via the increase in the nutrient digestibility, improvement in the rumen volatile fatty acids (VFA) composition, reduction in the ammonia nitrogen (NH_3-N) content as well as buffered rumen pH, and stimulation the growth rumen microbes [11,27,28]. However, exogenous fibrolytic enzymes, live yeasts, and yeast culture, among other biological methods for enhancing fiber utilization, have shown equivocal impacts on forage fiber digestion in individual experiments, but current meta-analyses indicate that there are overall benefits [29]. The increase in fiber digestion shown in current experiment may be explained by improving cell wall degradability and enabling ruminal bacteria to attack structural carbohydrates, resulting in enhanced hemicellulose and cellulose degradation as reported by Sun et al. [30].

Table 3 Dry matter intake and nutrient digestibility of swamp buffaloes fed with different types of treated rice straws.

Parameters	RS	UTS	YTS	ULTS	SEM	*p*-value
Intake						
g DM /d	5,600^{a}	5,620^{a}	7,150^{b}	7,090^{b}	25.697	0.029
%BW	1.90	1.99	2.21	2.11	0.180	0.086
g/kg BW$^{0.75}$	94.41^{a}	94.54^{a}	110.18^{b}	109.95^{b}	6.601	0.012
Digestion coefficient, %						
DM	54.36^{a}	63.43^{b}	68.38^{c}	64.75^{b}	0.158	0.033
OM	57.79^{a}	66.36^{b}	69.89 ^{c}	67.97^{b}	0.242	0.042
CP	50.73^{a}	60.01^{b}	62.43^{d}	61.32^{c}	0.305	0.038
NDF	50.35^{a}	55.98^{b}	58.99^{d}	57.97^{c}	0.425	0.031
ADF	44.42^{a}	51.66^{b}	54.47^{c}	52.65^{b}	0.238	0.037

RS = untreated, UTS = 2% urea, YTS = yeast, ULTS = 2% urea + 2% lime.
$^{a-d}$ Within the same rows, the means with different letters differ significantly ($p< 0.05$).

3.3 Rumen fermentation characteristics

Different types of treated rice straw affected the fermentation of rumen in buffalo (Table 4). No significant change in rumen pH between among groups ($p>0.05$) was observed, which was within the normal range of 6.6 to 6.9. Rumen NH_3-N levels in treated groups were greater than those in untreated groups ($p<0.05$). Furthermore, the yeast and urea-calcium hydroxide treatments led to the significant increase in the TVFA and C3 ($p<0.05$) contents, whereas no difference for C2 and C4 was observed in any of the groups ($p> 0.05$). The lower C2:C3 ratio in the rumen fluid of animals that were fed with all types of treated rice straw was related to the change in the molar proportion of C3. Rice straw treatment did not affect the rumen pH of the swamp buffaloes. In terms of microbial fiber digestion, all treatments remained within the normal range (6.3–7.0). The net effects of increased rumen pH probably promoted the activity of rumen bacteria, especially rumen cellulolytic bacteria, which exhibit extreme sensitivity to pH, and to increase rice straw digestibility and intake. Rumen ammonia nitrogen (NH_3-N) has long been known to be a key nutrient for rumen microorganisms. Rumen NH_3-N in the YTS and ULTS groups were greater than in the control group ($p<0.05$), and this result was similar to that reported by Wanapat and Pimpa [31]. Moreover, Wanapat and Rolinson [32] pointed that ruminal ammonia nitrogen (NH_3-N) is a key nutrient in rumen fermentation, with values ranging from 6.7 to 17.7 mg % depending on feeding conditions, and increasing ruminal NH3-N from 1.7 to 5.6 mg percent improved overall bacterial count as well as the digestibilities of DM, NDF, and ADF. Wanapat et al. [33] have demonstrated that swamp buffaloes are more efficient than cattle in terms of fiber digestion and rumen NH_3-N levels in terms of efficient fermentation and dry matter intake. Availability of energy, total nitrogen, and performed protein of rumen bacteria are key factors in the production of rumen microbial protein synthesis [34,35,36]. Total VFA concentrations and molar individual VFA were not significantly different with treated rice straw. Furthermore, Wanapat et al. [15] have reported that rumen VFAs are not affected by energy sources or urea levels. Nair et al. [37] have reported that C2 is reduced in dairy heifer that are fed corn silage inoculated with *S. cerevisiae*, whereas TVFA and C3 increase in dairy cows that are fed *S. cerevisiae* [38]. The molar proportion of C3 is changed, resulting in an increase in a potential glucogenic material for ruminants.

Table 4 Ruminal fermentation parameters of swamp buffalo fed with rice straw treated with different treatments.

Parameters	RS	UTS	YTS	ULTS	SEM	*p*-value
Rumen pH	6.6	6.7	6.7	6.9	0.155	0.125
Rumen NH_3-N, mg%	9.54^a	12.32^b	18.51^c	16.09^c	0.829	0.021
Total VFA, mmol/L	95.45^a	102.42^b	114.58^c	113.12^c	6.194	0.415
VFA, mol/100 mol						
Acetate, C2	73.78	71.61	70.05	70.76	1.391	0.862
Propionate, C3	17.89^a	20.32^b	22.64^c	21.39^{bc}	0.620	0.048
Butyrate, C4	8.33	8.07	7.31	7.85	0.377	0.106
C2:C3	4.12^a	3.52^b	3.09^c	3.31^{bc}	0.151	0.041

RS = untreated, UTS = 2% urea, YTS = yeast, ULTS = 2% urea + 2% lime.
$^{a\text{-}c}$ Within the same rows, the means with different letters differ significantly (p <0.05).

3.4 Rumen cellulolytic bacteria community

Table 5 shows the effect of different types of treated rice straw on the rumen microorganism population in buffaloes. Across all treatments, the predominant fibrolytic bacteria differed significantly, with the highest levels observed for yeast-treated rice straw (p<0.05). Predominant cellulolytic bacteria such as *F. succinogenes*, *R. flavefaciens*, and *R. albus* ranged from 2.35×10^{12} to 2.80×10^{12}, 0.74×10^{10} to 1.30×10^{10}, 0.78×10^{10} to 2.62×10^{10} copies/mL of rumen content, respectively. The population of *F. succinogenes* was the most abundant across dietary treatments. Population of cellulolytic bacteria was increased from 0-4 h post feeding. These results have been reported previously by our group [12], which are also consistent with that reported by Koike and Kobayashi [20] and Wanapat and Cherdthong [13] which found that *F. succinogenes* is the most abundant cellulolytic species in the swamp buffalo rumen, followed by *R. flavefaciens* and *R. albus*. Clearly, because *F. succinogenes* and *R. flavefaciens* can colonize cellulose more rapidly than the other two species, and it can grow more rapidly on cellodextrins (intermediate products obtained from cellulose hydrolysis). *R. albus* was regularly less widespread than *F. succinogenes* and *R. flavefaciens* because it was less efficient in cellulose colonization and hydrolysis than other species [12,39,40]. In addition, this result can be explained by the fact that yeast-treated rice straw which can provide sufficiently soluble growth factors for rumen microbes, leading to increased fiber degradation; the increased fiber digestion can subsequently lead to the increase in the feed intake and average daily gain. Yeast also has been related to increased fiber digestion as well as microbial protein flow and the activation of cellulolytic bacteria in the rumen.

Table 5 Quantification of rumen bacteria using real-time PCR as affected by rice straw treatments.

DNA copies of microbes	RS	UTS	YTS	ULTS	SEM	*p*-value
Total bacteria, $\times 10^{12}$/mL	$3.72^b \pm 0.52$	$4.09^b \pm 0.72$	$5.39^a \pm 0.15$	$5.23^a \pm 0.13$	0.098	0.040
F. succinogenes, $\times 10^{12}$/mL	2.35 ± 0.84	2.67 ± 0.90	2.80 ± 0.01	2.39 ± 0.35	0.418	0.138
R. albus, $\times 10^{10}$/mL	$0.78^b \pm 0.27$	$0.62^b \pm 0.24$	$2.62^a \pm 0.21$	$0.59^b \pm 0.08$	0.250	<0.001
R. flavefaciens, $\times 10^{10}$/mL	$1.16^a \pm 0.74$	$1.30^a \pm 0.47$	$0.89^b \pm 0.09$	$0.74^b \pm 0.18$	0.142	<0.001

RS = untreated, UTS = 2% urea, YTS = yeast, ULTS = 2% urea + 2% lime.
$^{a\text{-}b}$Within the same rows, the means with different letters differ significantly (p<0.05).

4. Conclusion

Based on this experiment, buffaloes that were fed yeast-treated rice straw exhibited increased DM feed intake, nutrient digestibility, improving fermentation efficiency of the rumen, and rumen population of cellulolytic bacteria. The efficiency of yeast-treated rice straw was greater than that of urea or urea-lime treated rice straw. Further studies in terms of meat and milk quality and feeding rice straw with fermented yeast could be conducted, which a good option to meet the increasing demand for feed resources with high potential utilization and a low environmental effect for buffalo production.

5. Ethical approval

The study was conducted according to the Kasetsart University Guide for the Care and Use of Animal (Protocol No. ACUC-KU 004/2019 provided by the corresponding author).

6. Acknowledgements

The authors acknowledge the administrative, financial and on-farm support provided by Department of Agricultural and Resources, Faculty of Natural Resources and Agro-Industry, Kasetsart University, Chalermphrakiat Sakon Nakhon Province Campus, Thailand for carrying out this study.

7. References

[1] Chantalakhana C. Urgent need in buffalo development for food security and self-sufficiency [Internet]. Göttingen: Cuvillier Verlag; 2001 [cited 2021 Mar 18]. Available from: http://hostcambodia.com/mekarn/procbuf/chan.htm.

[2] Devendra C. Perspectives on animal production systems in Asia. Livest Sci. 2007;106(1):1-18.

[3] Nada AS, Nakao T. Role of buffalo in the socioeconomic development of rural Asia: current status and future prospectus. Anim Sci J. 2003;74(6):443-455.

[4] Infascelli F, Gigli S, Campanile G. Buffalo meat production: performance infra vitam and quality of meat. Vet Res Commun. 2004;28 Suppl 1:143-148.

[5] Wanapat M, Ngarmsang A, Korkhuntot S, Nontaso N, Wachirapakorn C, Beakes G, et al. A comparative study on the rumen microbial population of cattle and swamp buffalo raised under traditional village conditions in the northeast of Thailand. Asian-australus J Anim Sci. 2000(7);13:918-921.

[6] Wanapat M, Sungchhang K, Phesatcha K. Enhancing buffalo production efficiency through rumen manipulation and nutrition. Buffalo Bull. 2013;32(1):258-275.

[7] Wanapat M, Chanthakhoun V, Pilajun R, Khejornsart P. Feed resources, rumen fermentation, manipulation and production in swamp buffalo: a review. In: Presicce PA, editor. The buffaloes (*Bubalus bubalis*)- production and research. 1st ed. Sharjah: Bentham Science Publishers Ltd; 2017. p. 145-179.

[8] Van Soest, PJ. Rice straw, the role of silica and treatments to improve quality. Anim Feed Sci Technol. 2006;130(130):137-171.

[9] Khejornsart P, Wanapat M. Effect of various chemical treated-rice straws on rumen fermentation characteristic using in vitro gas production technique. Livest Res Rural Dev. 2011;23(1):1-6.

[10] Wanapat M. Potential uses of local feed resources for ruminants. Trop Anim Health Prod. 2009;41(7):1035-1049.

[11] Wang Z, He Z, Beauchemin KA, Tang S, Zhou C, Han X, et al. Evaluation of different yeast species for improving in vitro fermentation of cereal straws. Asian- Australas J Anim Sci. 2016;29(2):230-240.

[12] Khejornsart P, Wanapat M, Rowlinson P. Diversity of anaerobic fungi and rumen fermentation characteristic in swamp buffalo and beef cattle fed different diets. Livestock Sci. 2011;139(3):230-236.

[13] Wanapat M, Cherdthong A. Use of real-time PCR technique in studying rumen cellulolytic bacteria population as affected by level of roughage in swamp buffalo. Curr Microbiol. 2009;58(4):294-299.

[14] Khejornsart P, Wanapat M. Effect of chemical treatment of rice straw on rumen fermentation characteristic, anerobic fungi diversity in vitro. J Anim Vet Adv. 2010;9(24):3070-3076.

[15] Wanapat M, Polyorach S, Boonnop K, Mapato C, Cherdthong A. Effect of treating rice straw with urea or urea and calcium hydroxide upon intake, digestibility, rumen fermentation and milk yield of dairy cows. Livest Sci. 2009;125(2):238-243.

[16] Latimer GW. Official methods of analysis of AOAC International. 19th ed. Gaithersburg: AOAC International; 2012.

[17] Soest VPJ, Robertso JB, Lewis BA. Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharides in relation to animal nutrition. J Dairy Sci. 1991;74(10):3583-3597.

[18] Keulen VJ, Young BA. Evaluation of acid insoluble ash as a neutral marker in ruminant digestibility studies. J Anim Sci. 1977;44(2):282-287.

[19] Yu Z, Morrison M. Improved extraction of PCR-quality community DNA from digesta and fecal samples. BioTechniques. 2004;36(5):808-812.

[20] Koike S, Kobayashi Y. Development and use of competitive PCR assays for the rumen cellulolytic bacteria: *Fibrobacter succinogenes*, *Ruminococcus albus* and *Ruminococcus flavefaciens*. FEMS Microbiol Lett 2001;204(2):361-366.

[21] Steel RGD, Torrie JH. Principles and procedures of statistics: a biometrical approach. 2nd ed. New York: McGraw-Hill Book Company; 1980.

[22] Shen HS, Sundstøl F, Eng ER, Eik LO. Studies on untreated and urea-treated rice straw from three cultivation seasons: 3. Histological investigations by light and scanning electron microscopy. Anim Feed Sci Tech. 1999;80(2):151-159.

[23] Man NV, Wiktorsson H. The effect of replacing grass with urea treated fresh rice straw in dairy cow diet. Asian Aust J Anim Sci. 2001;14(8):1090-1097.

[24] Zhang X, Rodolfo MF, Min W, Beauchemin KA, Zhiyuan M, Rong W, et al. Effects of urea plus nitrate pretreated rice straw and corn oil supplementation on fiber digestibility, nitrogen balance, rumen fermentation, microbiota and methane emissions in goats. J Anim Sci Bio. 2019;10(6):1-10.

[25] Wanapat M, Kang S, Hankla N, Phesatcha K. Effect of rice straw treatment on feed intake, rumen fermentation and milk production in lactating dairy cows. Afri J Agri Res. 2013;8(17):1677-1687.

[26] Trach NX, Mo M, Dan CX. Effects of treatment of rice straw with lime and/or urea on its chemical composition, in-vitro gas production and in-sacco degradation characteristics. Livest Res Rur Dev. 2001; 13(4):5-12.

[27] Durand CF, Ameilbonne A, Auret P, Bernard M, Mialon MM, Duniere L, et al. Supplementation of live yeast based feed additive in early life promotes rumen microbial colonization and fibrolytic potential in lambs. Sci Rep. 2019;9:19216.

[28] Campanile G, Zicarelli F, Vecchio D, Pacelli C, Neglia G, Balestrieri A, et al. Effects of *Saccharomyces cerevisiae* on in vivo organic matter digestibility and milk yield in buffalo cows. Livest Sci 2008;114(2): 358-361.

[29] Adesogan AT, Arriola KG, Jiang Y, Oyebade A, Paula EM, Cervantes PA, et al. Symposium review: technologies for improving fiber utilization. J Dairy Sci. 2019;102(6):5726-5755.

[30] Sun R, Lawther JM, Banks WB. Influence of alkaline pre-treatments on the cell wall components of wheat straw. Ind Crops Prod. 1995;4(2):127-145.

[31] Wanapat M, Pimpa O. Effect of ruminant NH3-N levels on ruminal fermentation, purine derivative, digestibility and rice straw intake in swamp buffalo. Asian-Aust J Anim Sci. 1999;12(6):904-907.

[32] Wanapat M, Rowlinson P. Nutrition and feeding of swamp buffalo: feed resources and rumen approach. Italian J Anim Sci. 2007;6(2):67-73.

[33] Wanapat M, Ngarmsang A, Korkhuntot S, Nontaso N, Wachirapakorn C, Beakes G, et al. A comparative study on the rumen microbial population of cattle and swamp buffalo raised under traditional village conditions in the northeast of Thailand. Asian-Aust J Anim Sci. 2000;13(7):478-482.

[34] Firkins J, Yu LZ, Morrison M. Ruminal nitrogen metabolism: perspectives for integration of microbiology and nutrition for dairy. J Dairy Sci. 2007;90 Suppl 1:E1-E16.

[35] Cherdthong A, Wanapat M, Kongmun P, Pilajan R, Khejornsart P. Rumen fermentation, microbial protein synthesis and cellulolytic bacterial population of swamp buffaloes as affected by roughage to concentrate ratio. J Anim Vet Adv. 2010;9(11):1667-1675.

[36] Hackmann TJ, Firkins JL. Maximizing efficiency of rumen microbial protein production. Front Microbiol. 2015;6:1-16.

[37] Nair J, Xu S, Smiley B, Yang HE, McAllister TA, Wang Y. Effects of inoculation of corn silage with *Lactobacillus* spp. or *Saccharomyces cerevisiae* alone or in combination on silage fermentation characteristics, nutrient digestibility, and growth performance of growing beef cattle. J Anim Sci. 2019;97(2):4974-4986.

[38] Bakr HA, Hassan M, Giadinis N, Panousis N, Andric´ DO, Tawab MA, et al. Effect of *Saccharomyces cerevisiae* supplementation on health and performance of dairy cows during transition and early lactation period. Biotechnol Anim Husb. 2015;31(3):349-364.

[39] Wanapat M, Pilajun R, Kongmun P. Ruminal ecology of swamp buffalo as influenced by dietary sources. J Anim Feed Sci Tech. 2009;151(3-4):205-214.

[40] Weimer PJ. Manipulating ruminal fermentation: a microbial ecological perspective. J Anim Sci. 1998;76(12):3114-3122.