

Lactation Performance of Nili-Ravi Buffaloes Fed Alkali Treated Rice Husks

Research Article

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ABSTRACT

A feeding trial was carried out using 20 lactating Nili-Ravi buffaloes, divided into 4 groups (n=5) having similar parity and milk production to examine the influence of 2% NaOH treated rice husk on their milk yield and composition, blood metabolites and nutrient digestibility. Four iso-nitrogenous (14.0% crude protein (CP)) and iso-caloric (metabolizable energy (ME) 2.5 Mcal/kg) rations, containing zero (control), 10%, 20% and 30% rice husk, were formulated. Buffaloes were individually fed the diets *ad libitum*. Ten kg of available green fodder (*Trifolium alexandrinum*, berseem) was also offered to each animal. Feed consumed was noted daily by subtracting refusals. Milking was twice a day with the yield being recorded. Milk samples were analyzed for fat, solids-not-fat, total solids and protein. A digestibility trial was conducted at the end of the study for seven days using three animals from each group. Milk yield and composition was similar across the treatments. Nutrient digestibility of dry matter, crude protein, crude fiber and ash increased linearly with increasing levels of treated rice husk, but differences were non-significant ($P>0.05$). Similarly, changes in blood metabolites also not significantly affected by the treatments. It can be concluded that treatment of rice husks with a low level of NaOH has no harmful effect, so can be used in the formulation of diets for lactating buffaloes.

KEY WORDS blood metabolites, buffalo, digestibility, milk production, rice husk.

INTRODUCTION

Production of good quality fodder and forage is one of the key factors in the development of a ruminant livestock industry as the performance of animals is directly related to fodder availability. Roughages constitute a major proportion of livestock feed in the developing countries. Crop residues (straws, stovers and hulls) are comparatively cheaper feed, but nutritive quality is lower than green forage or hay and, therefore, alone cannot sustain livestock production. Rice husk is one of the by-products from rice

processing, when the grain is removed from its fibrous covering, resulting in a widely available resource which is underutilized. The Food and Agriculture Organization of the United Nations (FAO, 2010) has estimated that more than 700 million tons of rice were produced in 2010. In rice cultivating regions large quantities of rice husk are disposed of as waste, or burned in open fields, thereby polluting the environment (Kim *et al.* 2003). The percentage of rice husk in paddy rice varies from 20-30% and is a cellulose-based fiber containing approximately 20% silica in an amorphous form.

Rice husk obtained from rice threshing has been used as an ingredient in ruminant and poultry feeds, but the problem of low nutrient digestibility, a high silica/ash content and an abrasiveness are limiting factors in its utilization.

Rice husk is composed of cellulose, hemicellulose and lignin and to break down these components, cellulase, hemicellulase and ligninase enzymes are required. These are not produced by the animals themselves, but the reticulo rumen of ruminants hosts the microorganisms that produce these enzymes. However, lignin cannot be broken down in the rumen due to the lack of ligninase, although lignin is important because of its role on nutrient degradability and feed intake. One component of the rice cell wall is silica, which can be present in large concentrations ranging from 5-15%, depending on the rice variety and the availability of this mineral in the soil (Agbagla-Dohnani *et al.* 2003). Silica reduces palatability and the degradability of rice straw in the rumen due to its direct action in preventing colonization by ruminal microorganisms.

To enhance digestibility and crude protein concentration, and to reduce silica content, physical and chemical treatments may be applied to the husks. Several studies have been reported on the physical and chemical treatment and utilization of both rice straw and husks as ruminant feed. In Their studies of rice straw, Selim *et al.* (2004) also used biological treatments and supplementing the diet.

Biochemical conversion of lignocellulosic biomass into value added products is a subject of great interest. For this to succeed, it is essential to alter the structure of the lignocellulosic biomass to make it more accessible to enzymes that convert carbohydrate polymers into fermentable sugars (Mosier and Wyman, 2005). Various chemical and biological treatments involve the alteration of biomass so that enzymatic hydrolysis of cellulose and hemicellulose can be achieved more rapidly and with greater yields (Mosier and Wyman, 2005).

The major effect of alkaline treatment of fibrous material is the removal of lignin, thus improving the reactivity of the remaining polysaccharides. In addition, alkali pre-treatments remove acetyl and the various uronic acid substitutions on hemicellulose that lower the accessibility of the enzyme to the hemicellulose and cellulose surface (Chang and Holtzapple, 2000). Usually lime (calcium hydroxide) or sodium hydroxide is used. By using these components, salts are formed that may be incorporated in the biomass and need to be removed or recycled (González and López-Santín, 1986).

Combined mechanical/alkaline treatment consists of a continuous mechanical treatment (e.g. milling, extrusion, refining) of the lignocellulosic biomass with the aid of an alkali. The resulting fractions consist of a soluble fraction (containing lignin, hemicellulose and inorganic compo-

nents) and a cellulose-enriched solid fraction (Chang and Holtzapple, 2000; Selim *et al.* 2004).

Based on reported evidence rice husk utilization in the current study was planned to improve its nutritional value through alkali treatment. Hence the study was conducted to: 1) measure the effects of alkali treatment; 2) examine digestibility of treated rice husk in lactating buffaloes and 3) assess treated rice husk, when added to a total mix ration (TMR), on milk production and its composition in lactating buffaloes.

MATERIALS AND METHODS

Treatment of rice husk

For the treatment of rice husk, 2%, 4% and 6% NaOH solutions were prepared in water and sprinkled thoroughly on to the rice husk. The treated rice husk was then packed in polyethylene bags (lab silos) and placed at room temperature in the lab for 15 days (Tauqir, 2010). After opening the lab silos, treated rice husk was analyzed chemically for proximate and fiber compositions (Table 1). Chemical composition of fodder *Trifolium alexandrinum* (berseem) is given in Table 2. On the basis of lab analysis, rice husk treated with 2% alkali was selected for preparation for the large scale trial with the lactating buffaloes.

Animals and diets

Twenty lactating buffaloes of similar body weight (group I- 602.17±9.82; group II- 583.65±18.11; group III- 625.31±34.39; group IV- 594.48±8.73) and in mid-stage of lactation with average group yield (group I- 9.98±1.49; group II- 9.71±1.92; group III- 9.62±1.37; group IV- 9.83±1.67) were selected from the herd maintained at the Livestock Experiment Station, Bahadurnagar, Okara. They were divided into 4 groups (5 buffaloes per group) according to a completely randomized design. Rice husk treated with 2% NaOH solution was used in the preparation of experimental rations. Four iso-caloric and iso-nitrogenous TMR diets were formulated using NRC (2001) standards for energy and protein. They contained 10, 20 or 30% alkali treated rice husks while the control ration contained 30% un-treated rice husks (Table 3).

Rations were provided *ad libitum* together with 10 kg available green fodder (berseem; *Trifolium alexandrinum*) to meet the vitamin and mineral contents and to enhance nutrient digestibility. Animals were individual fed. The trial lasted for 60 days, after a additional 7 day period for adaptation to the diets. Feed offered, refusals and milk production were recorded daily. Milk samples were taken weekly for the determination of fat, solids-not-fat (SNF), total solids and protein by using lacto scanner (Ekomilk Lacto Scan).

Table 1 Chemical composition (%) of treated & un-treated rice husk

Composition	Untreated	Treated
Organic matter	77.94	88.07
Dry matter	98.94	98.55
Crude protein	1.70	1.6
Neutral detergent fiber	78.82	69.03
Acid detergent fiber	70.14	66.22
Lignin	28.70	25.79
Ash	22.53	21.86
Metabolizable energy (MJ/Kg DM)	1.68	1.93

Table 2 Chemical composition (%) of berseem fodder

Berseem fodder	Chemical composition
Dry matter %	17
Organic matter %	92.2
Crude protein %	17
Neutral derangement fiber %	46.5
Acid derangement fiber %	29
Hemicellulose %	17.5
Cellulose %	10.6
Acid derangement lignin %	7
Ash %	11.4
Net energy (lactation)	1.45

Table 3 Ingredients and chemical composition of experimental rations¹

Ingredients %	Control	RH10	RH20	RH30
Maize broken	15.00	20.00	20.00	15.00
Rice husk	30.00	0.00	0.00	0.00
Rice husk alkali treated	0.00	10.00	20.00	30.00
Wheat bran	4.00	24.00	12.00	4.00
Cotton seed cake	4.50	4.50	4.50	4.50
Maize gluten meal 30%	20.00	20.00	20.00	20.00
Canola meal	10.00	5.00	7.00	10.00
Cane molasses	15.00	15.00	15.00	15.00
Mineral mix	1.50	1.50	1.50	1.50
Total	100	100	100	100
Chemical composition %				
Dry matter	90.24	88.32	89.20	90.09
Crude protein	13.57	14.90	14.03	13.54
Metabolizable energy (Mcal/kg)	2.47	2.68	2.63	2.55
Neutral detergent fiber	39.17	29.92	32.21	36.24
Acid detergent fiber	26.71	14.53	19.76	25.53
Acid detergent lignin	10.45	4.69	7.03	9.57
Cellulose	15.27	9.98	12.39	15.27
Hemicellulose	23.06	19.50	20.70	23.06
Ash	13.49	10.15	11.75	13.28

¹ Control ration contained 30% untreated rice husk, while RH10, RH20 and RH30 contained 10%, 20% and 30% alkali treated rice husk, respectively.

Digestibility trial

At the end of study, a digestibility trial was conducted using 3 animals from each group. Complete collection of feces was undertaken for 7 days. After weighing and composting, representative samples were stored in a freezer for further analysis. Data of daily TMR intakes was recorded separately by subtracting refusals from the amount offered. Diets, orts and fecal samples were analyzed for DM and CP (AOAC, 1990). (Proximate composition of fodder is pre-

sented as Table 2)

Blood collection

During the digestibility trial, 10 milliliters of blood was collected 3 hours post feeding from the jugular vein into a vacutainer for further examination. Serum was extracted by centrifuging it at 3500 rpm for 15 minutes. Blood urea nitrogen was determined according to the method prescribed by Bull *et al.* (1991), while blood glucose was determined

by using the crescent diagnostic glucose enzymatic colorimetric god-pap method (Trinder, 1969).

Statistical analysis

Data for feed intake, milk production and composition, nutrient digestibilities and blood metabolites was analyzed statistically using SPSS (2011) software and means were compared using Duncan's multiple range test (Steel *et al.* 1997). The statistical model used for all parameters was:

$$Y_{ij} = \mu + \beta_k + \tau_j + \varepsilon_{ijk}$$

Where:

μ : overall mean.

β_k and τ_j : effect of block and treatment (4 treatments) respectively.

ε_{ijk} : difference within treatments (error term).

RESULTS AND DISCUSSION

Intake of dry matter intake (DMI) and fiber (NDF) was the highest in animals fed the diet with 30% untreated rice husk. With treated rice husk, animals fed RH20 consumed more than those fed RH10 and RH30 (Table 4). The DMI showed a linear and cubic trend in the current study while DMI% of body weight also exhibited the similar trend. The intake of CP was not affected by treatment.

Feed intake was the highest in animals fed untreated rice husk compared to those receiving treated husks. This may be due to odor/flavor, taste or chemical changes occurring in rice husk due to lime treatment. Results of the current study have supported the findings of Cameron *et al.* (1990) who described a linear decrease in DM intake ($p=0.13$) when the amount of alkaline hydrogen peroxide treated wheat straw (AHP-WS) was increased in the diet. Dry matter intake for the ruminally cannulated cows was not different ($P=0.20$) among the diets. Consumption of DM as % of BW, decreased linearly ($P=0.15$) from 3.48% for the control to 3.13% for the high AHP-WS diet. It appeared that increasing AHP-WS in the diet of cows during early lactation slightly decreases DM intake. Other studies (Greenhalgh *et al.* 1976; Canale *et al.* 1988) reported that NaOH treatment of low-quality roughages resulted in an improved DM intake and milk yield when compared with control diets containing the untreated roughages. Intakes of DM, crude protein (CP) and neutral detergent fiber (NDF) were not affected by increasing AHP-WS in the diet. Acid detergent fiber intake increased linearly ($P=0.03$) from 3.7 kg/d for the control cows to 4.0, 4.5 and 4.9 kg/d for cows fed the low, medium and high AHP-WS diets, respectively, while a 32% increase between the control and the high AHP-WS diet was observed. Similarly, Duckworth (2013)

found that calcium oxide treatment of both modified wet distillers' grains with solubles and corn stover reduced ($P \leq 0.03$) DMI and final body weight when compared to feeding untreated corn stover. In contrast to this, Shreck *et al.* (2014) fed CaO treated (5% of DM) wheat straw and corn stover with wet distillers' grains plus solubles (WDGS) to 460 steer calves during a 69-day growth study and observed greater DMI and an improved feed to gain ratio compared to those fed corn stover diets. Alkaline treatment increased DMI and improved feed to gain ratio. Mixing of WDGS into CaO treated wheat straw before feeding reduced the negative effects of chemical changes occurred in corn stover silage due to lime treatment.

Sewell *et al.* (2009) studied nutrient digestion and performance of lambs and steers fed thermochemically treated crop residues and reported this benefited in nutrient digestion and subsequent animal performance. They further reported that processed crop residues may be fed in combination with distillers dried grains with solubles (DDGS) to partially replace corn in ruminant diets. Shreck *et al.* (2011) studied digestibility of crop residues after chemical treatment and anaerobic storage and reported improved digestibility.

It another study, Shreck *et al.* (2015) investigated digestibility and performance of steers fed low quality crop residues treated with calcium oxide to partially replace corn in finishing diets containing distillers grains. They concluded that the improvements in total tract fiber digestibility that occurred when treated forages were fed may have been related to increased digestibility of recoverable NDF and not to increased ruminal pH. Feeding chemically treated crop residues and WDGS is an effective strategy for replacing a portion of corn grain and roughage in feedlot diets.

Schroeder *et al.* (2014) studied the effects of calcium oxide treatment of dry and modified wet corn distillers grains plus solubles on growth, carcass characteristics and apparent digestibility of feedlot steers and found improved feed efficiency but no improvement in digestibility. In a recent study, Kume *et al.* (2019) reported that yam peel meal can be used to replace maize offal up to 100% without adverse effects on the growth performance and digestibility of red Sokoto goats.

Dry matter digestion was the highest in buffaloes fed the control diet and lowest in those fed the diet. It was similar in those in diet RH30 and lowest with the RH10 and RH20 diets (Table 4). Dry matter digestion increased linearly with increasing rice husk in the ration of lactating buffaloes. Dry matter and fiber digestibility trends were similar statistically. Crude protein digestion was similar in this study while fiber digestion displayed an analogous trend as was revealed in dry matter digestion (Table 4).

Table 4 Nutrient intake and their digestion in lactating buffaloes fed different level of treated and untreated rice husk¹

Parameters	Control	RH10	RH20	RH30	SEM	Linear	Quadratic	Cubic
Dry matter intake, kg/day	23.00 ^a	22.20 ^b	22.80 ^a	22.00 ^b	0.12	**	NS	**
DMI% body weight	3.86 ^{ab}	3.93 ^a	3.69 ^b	3.75 ^b	0.03	NS	NS	NS
Crude protein intake, kg/day	2.30	1.84	1.90	2.00	0.10	NS	NS	NS
Neutral detergent fiber intake, kg/day	9.12 ^a	6.85 ^d	7.42 ^c	8.08 ^b	0.19	**	**	**
Dry matter digestibility, %	60.20 ^{ab}	58.80 ^{bc}	58.00 ^{bc}	61.80 ^b	0.89	*	NS	NS
Crude protein digestibility, %	71.20	72.00	71.60	71.40	0.14	NS	NS	NS
Neutral detergent fiber digestibility, %	50.20 ^{ab}	49.80 ^{bc}	49.00 ^c	52.80 ^b	0.89	*	NS	NS

¹ Control ration contained 30% untreated rice husk, while RH10, RH20 and RH30 contained 10%, 20% and 30% alkali treated rice husk, respectively. The means within the same row with at least one common letter, do not have significant difference ($P > 0.05$).

** ($P \leq 0.01$) and * ($P \leq 0.05$).

SEM: standard error of the means.

NS: non significant.

Verma and Jaiswal (1981) studied the effect of NaOH treated wheat straw in Sahiwal steers and reported significantly better fiber digestibility when it was added to the ration. Rehman (1985) reported that an increase in the digestibility was similar when NaOH treated rice straw was fed to lactating buffaloes. Percent digestibility of crude fiber in non-lactating Nili-Ravi buffaloes was significantly affected by various treatments, while DM digestibility did not show any treatment effect (Kaur, 2010). It was concluded that NaOH treatment may be considered as more suitable in improving the nutritional value of crop residues because of its economics and practical viability.

Canale *et al.* (1988) reported an increased apparent digestibility of NDF, acid detergent fiber (ADF) and hemicellulose when Holstein cows were fed sodium hydroxide treated (4 g NaOH/100 g forage DM) alfalfa-orchard grass (23.4 vs. 22.2 kg/d). Intake of dry matter was also increased. The findings of Wanapat *et al.* (2009) revealed significant improvements in dry matter intake and digestibility from 5.5% urea-treated rice straw and 2.2% urea + 2.2% calcium hydroxide treated rice straw compared to untreated straw. Rumen pH and $\text{NH}_3\text{-N}$ levels were increased. The results of the present study depicted higher DM and fiber digestibility when low levels of alkali treated rice husk were added to the diets of buffaloes, and DM digestibility reflected fiber digestibility.

Milk production of animals fed the control, RH10 and RH30 diets were similar and better than those on RH20, but the difference were statistically non-significant (Table 5).

These results could have been more pronounced if the study period had been extended. Similar findings were also revealed with 4% fat corrected milk. Milk fat, solid-not-fat and total solids were similar from all the treatment groups (Table 5). It is important to note that the differences among the groups were not statistically significant, thus indicating a similar trend.

The results of the current study support the findings of Canale *et al.* (1988), who reported a non-significant intake of dry matter was greater when cows were fed treated hay (23.4 vs. 22.2 kg/d), as was milk yield (32.3 vs. 30.9 kg/d). Yield of 4% fat-corrected milk (FCM), however, did not differ between diets (27.8 vs. 27.4 kg/d). Cows fed the treated hay diet had increased concentrations of total rumen volatile fatty acids and ruminal acetate, decreased ruminal isobutyrate concentration and pH, and increased apparent digestibility of NDF, ADF, hemicellulose, and lignin. Sodium hydroxide treatment also increased the proportion of potentially digestible DM and NDF compared with that of untreated forage.

Derso (2009) observed increased milk yield from treating rice straw with urea: T1, 1.2 kg/cow/day (grazing+untreated rice straw); and 2.36, 2.48, and 2.63 kg/cow/day in T2 (grazing+urea treated rice straw), T3 (grazing+urea treated rice straw + rice bran) and T4 (grazing+urea treated rice straw+formulated concentrate mix), respectively. While, Doan *et al.* (1999) found a slight increase in milk production from 10.3 to 11.9 liters in Holstein-Friesian crossbred cows fed urea molasses block urea treated rice straw, respectively. Cameron *et al.* (1990) also conducted a study, with lactating Holstein-Friesian cows fed diets containing hydrogen peroxide treated wheat straw mixed with corn silage as fodder in different proportions, according to 4 × 4 latin square design. Milk yield, either as measured or corrected for 4% FCM was unaffected by treatment. Milk fat percentage increased (from 3.07 to 3.32%) and milk protein percentage decreased (from 2.61 to 2.56%) as the proportion of treated wheat straw in the diet increased. Cows fed the higher proportions of treated wheat straw had increased rumen concentrations of total VFA and molar percentage of acetate but a decreased molar percentage of propionate, resulting in an increased acetate to propionate ratio.

Table 5 Milk Production and its composition in lactating buffaloes fed different level of treated and untreated rice husk¹

Parameters	Control	RH10	RH20	RH30	SEM	Linear	Quadratic	Cubic
Milk, liters/day	9.25	9.55	7.88	9.56	0.52	NS	NS	NS
Milk fat, %	6.42	6.40	6.50	6.38	0.07	NS	NS	NS
4% fat corrected milk, liters/day	13.33	13.43	12.91	13.40	0.27	NS	NS	NS
Solid-not-fat, %	8.38	8.22	8.18	7.84	0.14	NS	NS	NS
Total solid, %	15.20	15.00	15.20	14.60	0.18	NS	NS	NS
Milk protein, %	3.65	3.62	3.68	3.70	0.06	NS	NS	NS

¹ Control ration contained 30% untreated rice husk, while RH10, RH20 and RH30 contained 10%, 20% and 30% alkali treated rice husk, respectively.

SEM: standard error of the means.

NS: non significant.

Table 6 Blood glucose and blood urea nitrogen (mg/dL) in lactating buffaloes fed different level of treated and untreated rice husk¹

Parameters	Control	RH10	RH20	RH30	SEM	Linear	Quadratic	Cubic
Blood glucose	18.60	19.00	22.60	19.20	0.81	NS	NS	NS
Blood urea nitrogen	46.80	41.80	46.60	44.20	1.95	NS	NS	NS

¹ Control ration contained 30% untreated rice husk, while RH10, RH20 and RH30 contained 10%, 20% and 30% alkali treated rice husk, respectively.

SEM: standard error of the means.

NS: non significant.

Contrary to our findings, [Wanapat et al. \(2009\)](#) observed 3.5% fat-corrected milk was highest; ranking from diet containing: 1, 2.2% urea + 2.2% calcium hydroxide treated rice straw; 2, 5.5% urea-treated rice straw and; 3, 2.2% urea-treated rice straw fed to groups in multiparous Holstein crossbred dairy cows.

This improvement in milk production may be due to the synergic effect of urea and calcium hydroxide treatment of rice straw. The combined effect not only increased protein contents but also improved energy availability through weakening/breaking the lignocellulosic bond. The use of a strong base mixed with a fibrous feedstuff has been shown to reduce the fiber fraction of high fiber, low quality feed stuffs to make them more digestible. Milk protein and fat concentrations were also increased.

Chemical treatment of poor quality roughage has the potential to increase nutrient digestibility and feed intake by the ruminant animal. Alkaline treatments disrupt the plant cell wall by dissolving some hemicelluloses and lignin and by swelling cellulose microfibrils. However, diets that contained NaOH-treated roughages fed to lactating dairy cows have resulted in varying effects on DM intake, milk yield, and milk composition ([Fayyaz et al. 2018](#)).

Blood urea nitrogen (BUN) was higher in the buffaloes fed RH20 compared to the other groups but the values were within the normal range of blood metabolites (Table 6). Blood glucose values also followed a similar trend as that observed in for BUN.

The blood glucose and blood urea nitrogen values did not show any treatment effect, although animals on the control and RH20 diets had higher values of blood glucose and blood urea nitrogen than other groups. It can be concluded that possibly buffalo of these two groups recycled BUN and blood glucose more efficiently, which were then available for the growth of fiber degrading bacteria resulting in improved performance.

Although the results were non-significant but when we extrapolate results or increase the trial period the trend may be clear as discussed above. Similar findings were reported by [Colovos et al. \(1967\)](#), who observed a non-significant effect of urea on blood glucose level. The findings of [Nadeem et al. \(2014\)](#) supported the present results, suggesting that blood urea nitrogen level was unaffected in Nili-Ravi buffaloes when urea was included in the diet. However, contrary to the current study findings, [Currier et al. \(2004\)](#) conducted a study on Angus and Hereford cows and observed the effect of urea supplementation on BUN level. It was increased with non-protein nitrogen (NPN) supplementation ($P < 0.01$) but not affected by NPN source or supplementation frequency.

CONCLUSION

The treatment of rice husk with 2% NaOH has no harmful effect so could be used in the formulation of diets for lactating buffaloes up to 20% without any discernable hazard. However more research is required to explore ways to improve its quality and value in animal feeding and its benefits on livestock productivity. Due to the prevalent situation of fodder shortages in term of quantity and quality especially during feed scarcity periods, techniques are needed that may applied to treat agro-industrial byproducts and that could be used for ruminant feeding in Pakistan. These results contribute to this and will be useful as a basis for further investigations in this field.

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