

Improvement of whole crop rice silage nutritive value and rumen degradability by molasses and urea supplementation

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Accepted: 5 June 2013 / Published online: 15 June 2013
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Abstract Whole crop rice was harvested 120 days after planting and chopped to 2–3-cm length for silage making. The whole crop rice silage (WCRS) was supplemented with different levels of molasses and urea to study nutritive value and in situ rumen degradability. The ensiling study was randomly assigned according to a 6×5 factorial arrangement, in which the first factor was molasses (M) supplementation at M0, M1, M2, M3, M4, and M5 %, and the second was urea (U) supplementation at U0, U0.5, U1.0, U1.5, and U2.0 % of the crop dry mater (DM), respectively. After 45 days of ensiling, temperature, pH, chemical composition, and fermentation end products of the silages were measured. Ten U and M treatment combinations of WCRS were subsequently selected to study rumen degradability by nylon bag technique. The results showed that temperature and pH of the silages linearly increased with U supplementation level, while total volatile fatty acid (TVFA), acetic acid (C2) and propionic acid (C3) decreased. In contrast, increasing level of M supplementation decreased WCRS temperature and pH, whereas TVFA, C2, and C3 concentrations increased dramatically. Both M and U supplementation increased concentration of butyric acid (C4). Dry matter, organic matter (OM), and acid detergent fiber (ADF) contents of the silages were not influenced by either M or U supplementation. Increasing U supplementation increased crude protein (CP) content, while M level did not show any effect. Furthermore, neutral detergent fiber (NDF) content in silage was decreased by both M and U supplementation. The results of the in situ study showed that M and U supplementation increased both ruminal DM and OM degradation. The water-soluble fraction (a) was the highest in WCRS U1.5M3

and lowest in U0M0. Increasing M and U supplementation levels increased the potentially degradable fraction (b) of both DM and OM. Total rumen degradable fraction (a+b) was highest in WCRS U1.5M3, whereas OM degradability was highest in U0M3. However, effective degradation of both DM and OM were the highest in WCRS U1.5M3 and the lowest in U0M0. We conclude that supplementation of U and M increases WCRS quality and rumen degradability. Supplementation of U at 1.5 and M at 3–4 % of the crop DM is recommended for lactating dairy cows and fattening beef cattle.

Keywords Whole crop rice · Silage · Urea · Molasses · Rumen degradability · Ruminant

Abbreviations

WCRS	Whole crop rice silage
DM	Dry matter
M	Molasses
U	Urea
CP	Crude protein
ADF	Acid detergent fiber
NDF	Neutral detergent fiber
OM	Organic matter
TVFA	Total volatile fatty acid
C2	Acetic acid
C3	Propionic acid
C4	Butyric acid

Introduction

In most parts of the world, forage conservation is a key element for productive and efficient ruminant livestock production. Forage conservation also provides farmers with means of preserving forage when production exceeds that of grazing animals. Silage making increased considerably from

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the 1960s and is the predominant method of forage preservation in temperate areas of the world (Cheli et al. 2013). Silage is widely used in farms and has a substantial role in animal production systems. High silage quality is a key factor to minimize cost of production and sustained animal health. Increasing use of silage has resulted in continuing efforts to minimize the quality losses (Bartzanas et al. 2013; Dunièrea et al. 2013). The primary goal of making silage is to maximize the preservation of original nutrients in the forage crop for feeding, usually a difficult task, since the fermentation process usually leads to less than optimal preservation of nutrients. The main aim of ensiling is to preserve fodder under anaerobic condition, where anaerobic microbes build up organic acids mainly lactic acid by using fermentable carbohydrates, and aerobic stability describes the length of time that silage remains stable (Mohd-Setapar et al. 2012; Seppälä et al. 2013).

Ensiling process is defined as involving the following steps: harvesting the crop at the optimal stage of maturity, chopping, loading into a silo, compacting and sealing to exclude air, storing, and finally unloading for animal feeding (Dunièrea et al. 2013). Ensiling, a preservation method for moist crops, is based on anaerobic fermentation, whereby epiphytic lactic acid bacteria convert water-soluble carbohydrates into organic acids, mainly lactic. As a result, the pH decreases, and the forage is preserved (Weinberg et al. 2010). Various different plant materials can be used for ensiling: grass, clover, alfalfa, barley, corn, wheat, sorghum, and various moist “by-products” of the food industry, such as apple pomace, beet pulp, and brewer's mash (Ajila et al. 2012) and have been used to feed dairy and beef animals in both developed and developing countries (Touqir et al. 2007a, b). Rice (*Oryza sativa* L. *Japonica*) is the main cash crop in Southeast Asian countries. Attention has been recently paid to rice whole crop as a new feed resource. The utilization of whole crop rice (WCR) as an animal feed has proven economically viable, not only as a way of disposing of rice straw residues but also as a real alternative for feeding livestock in regions, where rice is the main crop (Cao et al. 2009). Silage must be made from plant material with suitable moisture content, about 55–70%. Paddy rice is the main crop in Thailand, and rice crop have properties which can be used for silage making, since it contains 70.5 and 12.4% of moisture and CP, respectively.

Several processes have been developed to improve the fermentation and nutritional value of whole crop silage from forage paddy rice. Breeding programs are carried out, and newly developed rice varieties with increased yield and amount of digestible nutrients are being grown and tested. Also, harvesting, preparation, and storage techniques are constantly being improved. However, WCR is usually insufficient in sugars and lactic acid bacteria and may produce silages rich in ethanol rather than lactic acid and volatile fatty acid (VFA) (Cao et al. 2009). During the ensilage process, some bacteria are able to break down cellulose and hemicelluloses to various

simple sugars. Other bacteria break down simple sugars to smaller end products (acetic, lactic, and butyric acids). The most desirable end products are acetic and lactic acid. As the bacteria degrade starches and sugars to acetic and lactic acid, dry matter is lost. As bacteria consume sugars, end products produced, as it is the most efficient fermentation acid and will drop the pH of the silage the fastest. The faster the fermentation is completed, the more nutrients will be retained in the silage. Addition of carbon and nitrogen sources could improve the quality of silage. Therefore, the aim of this study was to investigate the effect of molasses and urea supplementation on whole crop rice silage quality and rumen degradation.

Materials and methods

Whole crop rice silage

Whole crop rice was harvested 120 days and immediately chopped to 2–3-cm length and ensiled to the respective supplementation treatments according to a 6×5 factorial arrangements in CRD. Factor A was molasses (M) supplementation at 0, 1, 2, 3, 4, and 5%, and factor B was urea (U) supplementation at 0, 0.5, 1.0, 1.5, and 2.0% of the crop DM. Whole crop rice was mixed with M and U and then packed into plastic bags. The silage bags were kept in ambient temperature (about 25–30 °C). All treatment combinations were done in triplicates at 1 kg each. After 45 days of ensiling, temperature and pH of silage were measured, and 200 g of whole crop rice silage were sampled for later chemical analysis. The samples were divided into two parts; first part was for DM analysis and second part was dried at 60 °C for 48 h and ground to pass through a 1-mm sieve in the feed mill using Cyclotec Mill, Tecator, Sweden for analysis of DM, OM and CP (AOAC 1990), and ADF and NDF (Van Soest et al. 1991).

In addition, silage was rinsed with distilled water and collected for volatile fatty acid analysis by using high-pressure liquid chromatography (Instruments by Water and Novapak model 600E; water mode I484 UV detector; column Novapak C18; column size 3.9×300 mm; mobile phase 10 mM H₂PO₄ [pH 2.5]) according to Samuel et al. (1997).

In situ trial

Ten treatment combinations were selected to study on rumen degradation by using nylon bag technique. Treatments were U0M0, U0M3, U0M4, U0M5, U1.5M3, U1.5M4, U1.5M5, U2M3, U2M4, and U2M5, where U is urea levels, and M is molasses levels. Samples were collected, oven-dried at 60 °C for 72 h, ground passed through a 1-mm screen and prepared for in Sacco analysis. Ruminal DM degradation of the feed fractions was determined by the nylon bag technique (Orskov and McDonald 1979). Bags, 8×14 cm in size with

a pore size of 40 μm , were used. Two grams of each sample treatments were placed into the bags and tied tightly with nylon thread. Two rumen fistulated swamp buffaloes, about 4 years old, with 440 ± 10 kg liveweight, were used in this study. All buffaloes were fed with rice straw ad libitum and supplemented with 0.5 % BW concentrate per day. All treatment bags were suspended in the rumen of swamp buffaloes. Incubation times (0, 3, 6, 9, 12, 15, 24, 48, and 72 h) were measured by placing the bags of each treatments into the rumen before the morning feeding and removals of two bags at each time. After removal, the bags were washed immediately in tap water until the water was clear and oven-dried at 60 °C for 72 h. Three bags of each treatment were washed under water without incubation in order to estimate losses at 0 h post feeding. Dry composited samples and the residues in the bag removed at the end of incubation time of treatments were measured for DM and organic matter (OM).

The ruminal disappearance characteristics of DM was fitted to the exponential following the procedure described by Orskov and McDonald (1979):

$P = a + b(1 - e^{-ct})$, where, p = disappearance (in percent) of DM and OM from the bags at the time t , a = proportion of immediately soluble fraction of DM and OM (in grams per kilogram), b = potentially but not immediately degradable fraction (in grams per kilogram), c = fractional rate of soluble degradation (in percent per hour) of b , and t is the incubation time (in hours). Solubility of DM and OM were determined similarly by washing of 0 h bags.

Data were fitted by using the NEWAY program (Chen 1996). The effective degradability of nutrients was calculated using the equation shown below, using a rumen out flow rate (k) of 0.02 per hour which is an average value for animals fed at approximately maintenance level (AFRC 1993): $ED = a + \{bc/(c+k)\}$.

Statistical analysis

All data were subjected to analysis of variance using Proc. ANOVA, and responding to level of urea and molasses were tested using orthogonal polynomial (SAS 1998). Dry matter and OM degradation means were statistically compared using Duncan's new multiple range test (Steel and Torrie 1980).

Results and discussion

Nutritive value of WCRS

Effect of M and U supplementation on WCRS characteristics and nutritive values are shown in Table 1. Temperature and pH of WCRS were increased, while total volatile fatty acid, acetic acid, and propionic acid concentration was decreased

linearly with increasing level of U. In contrast, supplementation of M decreased silage temperature, while no change of pH was found. Moreover, total volatile fatty acid (TVFA), acetic acid (C2), and propionic acid (C3) concentration were increased by M level supplementation. This was in contrast to the report of Alli et al. (1984) that silages were treated with M at a rate of 2.3 or 4.5 % fresh weight at the time of ensiling, which led to increased rates of lactic acid production, lower pH, and decreased DM loss. Lower pH value in silage could be due to higher lactic acid concentration in those silages. Cao et al. (2009) stated that lactic acid is the strongest of all silage acids, and its presence will drop pH more effectively than other VFA. The present study revealed that the result of pH was still above the ideal silage pH of 4.0 to 4.5. Moreover, proportion of C2/C3 was decreased with the increasing level of U supplementation in the silage, while M supplementation could increase the proportion of C2/C3. The VFAs comprises of C2, C3, butyric acid (C4), and other acids. The production of these acids is a reflection of an inefficient fermentation or of secondary fermentation of lactic acid to butyric acid and degradation of amino acids to ammonia with the production of acetic acid from the carbon skeleton of the amino acid (Santoso et al. 2011). Concentrations of TVFA, C2, and C3 were increased by M supplementation but not by U. This implied that M could improve the quality of silage. However, both U and M supplementation could increase the proportion of C4. The increased C4 in those silages could be due to higher pH (above 4.0 to 4.5) of the silage, which may favor the activity of clostridia. Santoso et al. (2011) stated that reducing pH silage prevented the growth of undesirable microbes, e.g., *Listeria*, clostridia, *Enterobacteriaceae*, and molds. However, it was found that interaction effect between M and U level was found on TVFA and proportion of C2/C3.

The greatest improvement in chemical composition was found in supplementation of U at 2 % and M at 4 % of the crop DM, which contained 15.2, 84.6, 63.7, and 47.2 % DM of CP, OM, NDF, and ADF, respectively. Based on this result, it showed that supplementation of M can improve WCRS quality, but not by U addition. Urea supplementation increased the CP content ($P < 0.01$). Moreover, both U and M supplementation decreased NDF content in the silage linearly. Molasses supplementation decreased DM and ADF content but U did not. However, OM, CP, and NDF were affected by interaction between U and M levels of supplementation. The nutritive values were relatively higher than the value of Ki et al. (2009), especially the CP content. Based on the findings of Khan et al. (2006) and Touqir et al. (2007a, b), who reported that increased concentrations of NDF and ADF but reduced CP in the silage were found when compared to whole rice crop, indicating the loss of CP during ensiling. This was similar to the present study that NDF and ADF content were increased by U supplementation; however, U level could increase the CP content in the present WCRS. On the other hand, M could reduce NDF and ADF content but

Table 1 Effect of molasses and urea supplementation on whole crop rice silage chemical composition and nutritive value

Items	Urea					SEM	Molasses						SEM
	0.0	0.5	1.0	1.5	2.0		0	1	2	3	4	5	
T, °C	27.6 ^c	28.0 ^{ab}	27.8 ^{bc}	28.2 ^a	28.2 ^a	0.12	28.0 ^x	27.9 ^{xy}	27.9 ^{xyz}	27.8 ^z	28.0 ^z	28.1 ^{yz}	0.13
pH	5.0 ^d	4.9 ^d	5.2 ^c	5.3 ^b	5.5 ^a	0.03	5.2	5.2	5.2	5.1	5.1	5.1	0.03
TVFA	189.8 ^a	182.9 ^{ab}	176.6 ^b	161.0 ^c	151.9 ^c	4.26	159.4 ^y	178.8 ^x	170.4 ^{xy}	168.4 ^{xy}	175.4 ^x	182.3 ^x	4.66
VFA, mol/100 mol													
C2	118.8 ^a	106.1 ^{bc}	110.0 ^b	96.2 ^d	100.2 ^{cd}	2.72	96.8 ^z	108.7 ^{xy}	106.1 ^{xy}	102.8 ^{yz}	109.3 ^{xy}	113.9 ^x	2.98
C3	56.0 ^{ab}	59.0 ^a	52.0 ^{bc}	48.2 ^c	38.6 ^d	1.73	48.1 ^y	54.7 ^x	50.2 ^{xy}	50.0 ^{xy}	49.8 ^{xy}	51.7 ^{xy}	1.90
C4	14.9 ^{bc}	17.8 ^a	14.7 ^{bc}	16.6 ^{ab}	13.2 ^c	0.69	14.6 ^{xy}	15.4 ^{xy}	14.2 ^y	15.6 ^{xy}	16.3 ^{xy}	16.7 ^x	0.76
Chemical composition													
DM, %	28.6	29.4	27.3	30.1	28.4	2.17	27.6	28.7	28.2	29.2	29.9	30.7	2.24
% of DM													
OM	92.3	94.3	96.2	94.7	95.6	1.84	94.5	95.3	94.6	95.7	94.1	96.4	1.87
CP	11.6 ^c	12.7 ^{bc}	13.5 ^{ab}	14.4 ^{ab}	15.2 ^a	0.61	13.2	13.7	13.8	13.1	14.5	13.7	0.65
NDF	65.2 ^{ab}	66.5 ^a	64.5 ^{ab}	64.0 ^{ab}	63.9 ^b	1.09	66.3 ^x	65.4 ^{xy}	64.9 ^{xy}	64.1 ^{xy}	63.2 ^y	63.7 ^{xy}	1.14
ADF	47.7	47.6	46.2	48.8	46.4	1.06	48.6	47.4	47.5	46.9	46.3	47.1	1.19

abc, xyz Mean with different superscripts in row ($P < 0.05$)

not CP. This indicated that combination of U and M supplementation could enhance the quality of the WCRS.

Dry matter and organic matter digestibility

Table 2 shows WCRS on ruminal DM and OM degradability. It was found that water-soluble fraction (a) was highest in U1.5M3 and lowest in U0M0 (23.8 and 14.6 %, respectively).

Potentially degradable fraction (b) differed among treatments ($P < 0.05$), which were the highest in U1.5M4 and lowest in U0M0 (55.6 and 45.0 %, respectively). Fractional rate of degradation was highest in U0M0 (0.025). Total rumen degradable fraction (a+b) and effective DM degradation similarly responded to treatments and were highest in U1.5M3 and lowest in U0M. Furthermore, although some researchers reported that adding M has no effect on DM digestibility

Table 2 Effect of urea and molasses supplementation on rumen degradability of whole crop rice silage

Item	Treatments ¹										SEM	
	U0M0	U0M3	U0M4	U0M5	U1.5 M3	U1.5 M4	U1.5 M5	U2M3	U2M4	U2M5		
DM digestibility												
a	14.6 ^a	20.4 ^{bc}	19.4 ^{abc}	18.5 ^{abc}	23.8 ^c	17.7 ^{ab}	21.1 ^{bc}	21.8 ^{bc}	19.5 ^{abc}	18.8 ^{bc}	1.69	
b	45.0 ^a	48.6 ^{ab}	53.0 ^c	53.4 ^c	53.0 ^c	55.6 ^c	52.4 ^{bc}	54.3 ^c	55.2 ^c	51.7 ^{bc}	1.34	
c	0.025 ^a	0.020 ^{ab}	0.019 ^{ab}	0.019 ^{ab}	0.017 ^b	0.019 ^{ab}	0.016 ^b	0.014 ^b	0.016 ^b	0.017 ^b	0.01	
a+b	59.6 ^a	69.0 ^{bc}	72.5 ^{bc}	71.9 ^b	76.8 ^c	73.3 ^{bc}	73.5 ^{bc}	76.1 ^{bc}	74.7 ^{bc}	70.5 ^{bc}	2.24	
ED	39.6 ^a	44.5 ^{bc}	44.9 ^{bc}	44.2 ^b	48.0 ^c	44.8 ^{bc}	44.2 ^b	43.9 ^b	43.9 ^b	42.5 ^{ab}	1.13	
OM digestibility												
a	10.6 ^{ab}	19.7 ^a	16.3 ^{ab}	14.2 ^{ab}	18.0 ^{ab}	9.0 ^b	15.1 ^{ab}	19.1 ^a	12.4 ^{ab}	13.2 ^{ab}	2.70	
b	51.0 ^{abc}	57.1 ^{ab}	59.3 ^a	44.3 ^c	47.7 ^{bc}	58.7 ^a	53.2 ^{abc}	49.9 ^{abc}	52.8 ^{abc}	48.4 ^{abc}	3.10	
c	0.023	0.017	0.021	0.035	0.037	0.042	0.028	0.018	0.030	0.027	0.01	
a+b	61.6 ^{ab}	76.8 ^a	75.6 ^a	58.6 ^b	65.8 ^{ab}	67.7 ^{ab}	68.4 ^{ab}	69.0 ^{ab}	65.2 ^{ab}	61.5 ^{ab}	4.36	
ED	37.3 ^a	45.6 ^{abc}	45.8 ^{bc}	42.4 ^{abc}	48.8 ^b	45.9 ^{bc}	43.9 ^{abc}	42.1 ^{abc}	42.9 ^{abc}	40.3 ^{ac}	2.02	

¹ U0, U1.5, and U2 were urea supplementation at 0, 1.5, and 2 % of the crop DM respectively, and M0, M3, M4, and M5 were molasses supplementation at 0, 3, 4, and 5 % of the crop DM, respectively

a Proportion of immediately soluble fraction of DM and OM (g/kg), b potentially but not immediately degradable fraction (g/kg), c fractional rate of soluble degradation (%/h) of b, and t is the incubation time (h), ED the effective degradability of nutrients

abc Mean with different superscripts in row different ($P < 0.05$)

(Granzin and Dryden 2005; Wang and Goetsch 1998, while others studies (Shellito et al. 2006; Sahoo and Walli 2008) have been reported that diets with M had higher ruminal DM digestibility. The OM disappearance (in percent) and estimated parameters of WCRS with different level of M and U are presented in Table 2. At all incubation times, there were significant ($P<0.05$) differences in OM disappearance of U0M4 significantly ($P<0.05$) higher than those supplemented. Therefore, supplementation of M and U at U0M4 had higher c, b, and ED than in other treatments. Organic matter disappearance could be decreased when M were increasing. However, U supplementation did not affect on OM digestibility.

Based on this study, it could be concluded that supplementation of U and M could efficiently increase whole crop rice silage quality and, thus, could provide good fermentation end product and chemical composition being high in protein and low in NDF contents. Present results suggested that supplementation of U and M to whole crop rice silage enhanced rumen degradation especially by addition U at 1.5 % and M at 3–4 % of the crop DM. Therefore, further study using whole crop rice silage supplemented with U and M in feeding trial emphasizing on lactating dairy cows and fattening beef cattle should be investigated.

Acknowledgment The authors are grateful for the financial support, research collaboration, and facilitations of the Tropical Feed Resources Research and Development Center (TROFREC), Department of Animal Science, Faculty of Agriculture, Khon Kaen University.

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