

## Influence of Ripening Stages on the Quality of Whole Crop Silage and Grain Silage of Fodder Rice

S. Maruyama<sup>1</sup>, I. Yokoyama<sup>2</sup>, H. Asai<sup>2</sup>, S. Sakaguchi<sup>2</sup>, T. Ohtani<sup>2</sup>, H. Yokota and K. Kita\*

Laboratory of Animal Feeds and Production, University Farm, Graduate School of Bioagricultural Sciences  
Nagoya University, Togo, Aichi 470-0151, Japan

**ABSTRACT :** In high-income Asian countries like Korea and Japan, per capita rice consumption has declined because of the change in consumer's favorite foods from rice to high-cost quality foods. This has forced farmers to reduce rice production. Although fodder rice could be another option to be cultured by farmers, available information concerning rice grain silage has been limited. In the present study, therefore, the difference in the quality of fodder rice silage prepared from either whole crop or grain at different ripening stages was compared. Various supplements were also added into whole crop and grain silages of fodder rice before ensiling, and thereafter, the palatability of prepared silages was determined by beef cattle. At ear emergence stage, the pH values for both grain and whole crop silages were approximately 4.5. In both grain and whole crop silages, the pH values were significantly increased by progressing ripening stages from milk-ripe stage to yellow-ripe stages, and the increase in pH value for grain silage was faster than that for whole crop silage. In the grain silage, the higher lactic acid (LA) content in grain silage seemed to be, the lower pH value was. Both in grain and whole crop silages, pH was significantly decreased by supplementation with LA bacteria. There were no significant differences in feed intake among any treatment groups. (*Asian-Aust. J. Anim. Sci. 2005. Vol 18, No. 3 : 340-344*)

**Key Words :** Fodder Rice, Whole Crop Silage, Grain Silage, Ripening, Silage Quality

### INTRODUCTION

It has been well recognized that rice is the staple food of Asia and part of the Pacific. Over 90 percent of the world's rice is produced and consumed in the Asia-Pacific Region. According to the report by Chaudhary (2000), during 10 years from 1987 to 1997 negative growth rate of rice area has been observed in Bangladesh, China, Fiji, Japan and Korea, and the decline of rice area would be resulted by less land, water, pesticide and labor in most countries. However, the situation of Korea and Japan was different from that in other countries. In Korea and Japan, income has been grown with growing prosperity and urbanization, and consumers substituted rice with high-cost quality food containing more protein and vitamins such as processed preparations of rice, vegetables, bread, fish and meat. This was the reason to lead the decline in per capita rice consumption especially in the middle and high-income Asian countries like Korea and Japan (Papademetriou, 2000).

In Japan, most of rice paddy field which had to be reduced was used for making other plants, for example wheat, soybean and sorghum, without rice. Another option of use of excess rice paddy field was aimed at making

fodder rice with more leaves than edible rice for food. Fukumi et al. (1984) compared several varieties of paddy rice to select the appropriate type as fodder rice. There has been several reports to investigate the quality of whole crop silage of fodder rice plant, so far (Nakui et al., 1985,1988; Yoshida et al., 1987; Enishi and Shijimaya, 1998). Compared to whole crop rice silage, however, the use of rice grain as silage material has been far limited. Nakui et al. (1986a) harvested rice grain at different ripening stages and attempted to prepare rice grain silage. In this report, carbohydrate content was increased by progressing ripening stages, but the contents of crude protein (CP) and neutral detergent fiber tended to be decreased. Nakui et al. (1986b) also reported the effect of supplementation of propionic acid (PA) and ammonia water on the prevention of aerobic deterioration of rice grain silage. Except for these reports, however, available information concerning with rice grain silage has not been reported. In the present study, therefore, we compared the difference in the quality of fodder rice silages prepared from either whole crop or grain at different ripening stages, and examined the palatability of the silages by beef cattle.

### MATERIALS AND METHODS

Two experiments were conducted. In experiment 1, the influence of different ripening stages on the quality of whole crop and grain silages of fodder rice was examined. In experiment 2, various supplements were added into whole crop and grain silages of fodder rice before ensiling,

\* Corresponding Author: K. Kita. Tel: +81-561-37-0203, Fax: +81-561-37-0203, E-mail: kitak@agr.nagoya-u.ac.jp

<sup>1</sup> Present address is Gifu Prefectural Livestock Research Institute, Kiyomi-mura, Ohno-gun, Gifu 506-0101, Japan.

<sup>2</sup> Gifu Prefectural Livestock Research Institute, Kiyomi-mura, Ohno-gun, Gifu 506-0101, Japan.

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**Table 1.** Ripening stages of fodder rice plant (*Oryza. sativa* L. Cv. Akichikara) which was transplanted on May 20, 1999

Date	Ripening stages	
August	17	Ear emergence stage
	24	Milk-ripe stage
	31	Dough-ripe stage
September	7	Yellow-ripe stage (prophase)
	14	Yellow-ripe stage
	21	Full-ripe stage

and thereafter, the palatability of prepared silages was determined by beef cattle.

### Silage preparation and palatability

In experiment 1, fodder rice seeds (*Oryza. sativa* L. Cv. Akichikara) were sown on April 23, 1999 and then seedlings were transplanted paddy rice field (Kokufu-cho, Gifu Prefecture, Japan) on May 20. Ripening stages of fodder rice were determined at the same quadrat (1.5 m × 1.5 m) every week from August, 17 to September 21. At the same time, fodder rice was also harvested for preparation of grain silage and whole crop silage. The whole crop was cut into 2 to 3 cm of length and ensiled in laboratory glass silo of 900 ml of volume. Grains were separated from whole crop and prepared grain silage like as whole crop silage. The laboratory silos containing whole crop or grains were stored at room temperature. The contents of dry matter (DM), CP, ether extract (EE), crude fiber (CF), ash and water soluble carbohydrate (WSC) in silage materials were measured. After 16 weeks of silage ensiling on August 17, silos were opened. Thereafter, pH and the contents of LA, volatile basic nitrogen (VBN), volatile fatty acids (VFA), acetic acid (AA), PA, iso-butyric acid (IBA), butyric acid (BA) and ethanol (ETH) were measured.

In experiment 2, fodder rice was cultured as described in experiment 1. Fodder rice at full-ripe stage was harvested on September 20. The 10 kg of whole crop or grains was ensiled in a nylon bag. To improve silage quality several supplements were added into whole crop silage and grain silage. Supplements used in the present study were cellulase (a mixture of *Acremonium cellulolyticus* and *Trichoderma viride* (1:2) which had 424 U/g of abicerase activity (Snow Brand Seed Co. Ltd., Sapporo, Japan) and LA bacteria (*Lactobacillus casei* sub sp. Rhamnusus, 10<sup>5</sup> cfc/g (Snow Brand Seed Co. Ltd., Sapporo, Japan). Water soaking treatment (24 h) was also done for rice grains. Nylon bags containing whole crop or rice grains were stored at room temperature. To investigate of palatability of silage, 27 female Japanese Black cattle (average BW 468 kg) reared in Gifu Prefectural Livestock Research Institute (Gifu, Japan) were used and divided evenly into 9 experimental groups of 3 cattle each. Before measuring the palatability, cattle were trained to feed silage from box trough for 1 week. The 3 kg each of silage was given to each 3 cattle.

Feed intake for 4 min was measured as the index of palatability of silage.

### Chemical analysis

The contents of DM, CP, EE CF and ash were measured by standard procedure (AOAC, 1990). CP in the ingredients of experimental diets was determined by using Kjeldahl distilling unit "Kjeltec System 1026" (Tecator, Hoganas, Sweden). EE was analyzed by Soxhlet's extractor "FATEX Speedy Fat Extractor Auto Program System" (Mitamura Riken Kogyo Inc., Tokyo, Japan). The WSC concentration was determined by the anthrone reaction rate assay (Koehler, 1952). The values for pH, LA and VBN in the silage were analyzed by the methods described by Ohshima et al. (1991). The profile of various VFA was analyzed by gas-chromatograph Shimadzu GC-12A (Kyoto, Japan). The ethanol concentration was determined by a commercial kit (Testkit Ethanol, R-Biopharm GmbH, Germany).

### Statistical analyses

Statistical analyses of data were performed by one-way ANOVA to assess the effects of ripening stage (experiment 1) and treatment (experiment 2). Then, Duncan's multiple range test was performed to compare all means. Two-way ANOVA was also performed to test main and interactive effects of ripening stage and silage material (experiment 1). All statistical analyses were performed using the General Linear Model Procedures (SAS Institute Inc., 1990). Differences between means were considered to be significant at  $p < 0.05$ .

## RESULTS

Ripening stages of fodder rice plant (*Oryza. sativa* L. Cv. Akichikara) which was transplanted on May 20, 1999 were described in Table 1. Briefly, ear emergence stage was August 17, milk-ripe stage was August 24, dough-ripe stage was August 31, yellow-ripe stage (prophase) was September 7, yellow-ripe stage was at September 14, and full-ripe stage was at September 21.

Chemical compositions of fodder rice plant material harvested at different ripening stages were shown in Table 2. The DM content of grain increased gradually from 41.0% to 73.3% with progressing ripening stages from ear emergence stage to yellow-ripe stage. The DM content in the whole crop also increased from 24.7% (ear emergence stage) to 45.2% (yellow-ripe stage). The DM content of grain was higher than that in the whole crop through all ripening stages. The CF contents of grain and whole crop silages ranged from 10.7% to 27.1% and from 21.4% to 32.6%, respectively. The CF content was decreased gradually with progressing ripening stages in both grain and whole crop. The values for CP, EE, ash and WSC in both grain and whole crop were almost constant through ripening stages.

**Table 2.** Chemical compositions of fodder rice plant material harvested on different ripening stage

Silage material	Ripening stages	DM (%)	CP (%DM)	EE (%DM)	CF (%DM)	Ash (%DM)	WSC (%DM)
Grain	Ear emergence stage	41.0	6.2	1.9	27.1	5.4	2.6
	Milk-ripe stage	51.1	6.7	2.3	15.1	4.3	1.5
	Dough-ripe stage	61.4	6.7	2.1	12.8	3.7	1.1
	Yellow-ripe stage (prophase)	64.9	5.7	2.1	11.5	4.0	0.9
	Yellow-ripe stage	73.3	6.1	2.2	10.7	4.1	1.0
	Full-ripe stage	71.7	5.7	1.9	11.4	4.3	0.7
Whole crop	Ear emergence stage	24.7	7.4	1.3	32.6	8.8	2.2
	Milk-ripe stage	31.2	5.9	1.3	30.5	8.9	2.0
	Dough-ripe stage	34.7	6.1	1.2	25.0	8.1	1.2
	Yellow-ripe stage (prophase)	39.9	5.0	1.4	25.8	8.5	0.9
	Yellow-ripe stage	45.2	5.0	1.4	23.1	7.2	2.3
	Full-ripe stage	41.9	6.0	2.1	21.4	6.6	1.8

DM: dry matter; CP: crude protein; EE: ether extract; CF: crude fiber; WSC: water soluble carbohydrate.

**Table 3.** Chemical compositions of grain silage and whole crop silage of fodder rice plant harvested on different ripening stages (Experiment 1)

Silage material	Ripening stages	pH	LA (%DM)	VBN (%TN)	AA (%DM)	PA (%DM)	IBA (%DM)	BA (%DM)	ETH (%DM)
Grain	Ear emergence stage	4.59 <sup>e</sup>	0.39 <sup>b</sup>	9.0 <sup>b</sup>	1.10 <sup>c</sup>	0.05 <sup>de</sup>	0.007 <sup>ab</sup>	0.91 <sup>b</sup>	3.60 <sup>a</sup>
	Milk-ripe stage	4.30 <sup>f</sup>	1.57 <sup>a</sup>	9.8 <sup>b</sup>	0.77 <sup>d</sup>	0.02 <sup>e</sup>	0.010 <sup>a</sup>	0.31 <sup>de</sup>	2.38 <sup>c</sup>
	Dough-ripe stage	5.26 <sup>c</sup>	0.10 <sup>bc</sup>	3.0 <sup>e</sup>	0.26 <sup>f</sup>	0.02 <sup>e</sup>	0.003 <sup>ab</sup>	0.32 <sup>de</sup>	2.28 <sup>cd</sup>
	Yellow-ripe stage (prophase)	5.24 <sup>c</sup>	0.27 <sup>bc</sup>	2.2 <sup>e</sup>	0.18 <sup>fg</sup>	0.01 <sup>e</sup>	0.000 <sup>b</sup>	0.06 <sup>e</sup>	1.75 <sup>de</sup>
	Yellow-ripe stage	6.59 <sup>a</sup>	0.00 <sup>c</sup>	0.8 <sup>f</sup>	0.00 <sup>g</sup>	0.00 <sup>e</sup>	0.000 <sup>b</sup>	0.00 <sup>e</sup>	1.17 <sup>e</sup>
	Full-ripe stage	6.58 <sup>a</sup>	0.04 <sup>c</sup>	0.8 <sup>f</sup>	0.00 <sup>g</sup>	0.00 <sup>e</sup>	0.000 <sup>b</sup>	0.00 <sup>e</sup>	1.19 <sup>e</sup>
Whole crop	Ear emergence stage	4.53 <sup>e</sup>	0.09 <sup>bc</sup>	12.5 <sup>a</sup>	3.26 <sup>a</sup>	0.18 <sup>a</sup>	0.000 <sup>b</sup>	2.46 <sup>a</sup>	2.88 <sup>bc</sup>
	Milk-ripe stage	4.54 <sup>e</sup>	0.14 <sup>bc</sup>	11.4 <sup>a</sup>	2.34 <sup>b</sup>	0.10 <sup>bc</sup>	0.007 <sup>ab</sup>	0.96 <sup>b</sup>	2.44 <sup>c</sup>
	Dough-ripe stage	4.66 <sup>e</sup>	0.41 <sup>b</sup>	7.9 <sup>c</sup>	1.26 <sup>c</sup>	0.13 <sup>b</sup>	0.010 <sup>a</sup>	0.89 <sup>b</sup>	2.31 <sup>cd</sup>
	Yellow-ripe stage (prophase)	5.04 <sup>d</sup>	0.22 <sup>bc</sup>	7.4 <sup>c</sup>	0.76 <sup>d</sup>	0.11 <sup>bc</sup>	0.007 <sup>ab</sup>	0.55 <sup>cd</sup>	2.34 <sup>cd</sup>
	Yellow-ripe stage	5.66 <sup>b</sup>	0.24 <sup>bc</sup>	5.7 <sup>d</sup>	0.37 <sup>ef</sup>	0.02 <sup>e</sup>	0.003 <sup>ab</sup>	0.19 <sup>e</sup>	3.27 <sup>ab</sup>
	Full-ripe stage	4.33 <sup>f</sup>	1.38 <sup>a</sup>	7.3 <sup>c</sup>	0.57 <sup>de</sup>	0.08 <sup>cd</sup>	0.007 <sup>ab</sup>	0.68 <sup>bc</sup>	3.05 <sup>ab</sup>
SEM		0.053	0.097	0.38	0.074	0.014	0.0024	0.11	0.19
Statistical significance (two-way ANOVA), P									
Silage material		0.001	0.754	0.001	0.001	0.001	0.116	0.001	0.001
Ripening stage		0.001	0.001	0.001	0.001	0.001	0.080	0.001	0.001
Interactions		0.001	0.001	0.001	0.001	0.014	0.028	0.001	0.001

DM: dry matter; LA: lactic acid; VBN: volatile basic nitrogen; TN: total nitrogen.

AA: acetic acid; PA: propionic acid; IBA: iso butyric acid; BA: butyric acid; ETH: ethanol.

Means not sharing with a common superscript letter are significantly different ( $p < 0.05$  Duncan's multiple range test,  $n=3$ ).

Two-way ANOVA was performed to test main and interactive effects of silage material and ripening stage.

Chemical compositions of grain and whole crop silages of fodder rice plant harvested at different ripening stages were shown in Table 3. There were significant interactions between silage material (grain and whole crop) and ripening stage on all parameters measured. At ear emergence stage, the pH values for both grain and whole crop silages were approximately 4.5. In both grain and whole crop silages, the pH values were significantly increased by progressing ripening stages from milk-ripe stage to yellow-ripe stage, and the increase in pH value for grain silage was faster than that for whole crop silage. LA content in grain silage seemed to be high when its pH showed low value. At ear emergence stage, the value for volatile VBN of grain and whole crop silages were 9.0 and 12.5, respectively. VBN in both silages was significantly decreased by progressing

ripening stages from dough-ripe stage to yellow-ripe stage, and the decrease in VBN of grain silage was faster than that of whole crop silage. In the grain silage, AA was detected at the level of 1.10% at ear emergence stage and decreased gradually. AA content of whole crop silage was 3.26% at ear emergence stage and also decreased to 0.57% at full-ripe stage. Throughout the experimental periods, AA content of grain silage was less than that of whole crop silage. PA, IBA and BA contents in both grain and whole crop silages were lower than LA and AA contents. ETH content of grain silage was decreased from 3.60% at ear emergence stage to 1.19% at full-ripe stage. Compared to grain silage, ETH content of whole crop silage remained constant during ripening stages.

The pH values in silages added with additives were

**Table 4.** Palatability of whole crop silage and grain silage of fodder rice plant material (Experiment 2)

Silage	Supplement	pH	Feed intake (kg/4 min)
Whole crop silage	None	4.60 <sup>cd</sup>	0.98
	Cellulase	4.54 <sup>de</sup>	0.71
	Lactic acid bacteria	4.45 <sup>ef</sup>	0.85
	Cellulase+lactic acid bacteria	4.50 <sup>def</sup>	0.98
Grain silage	None	6.73 <sup>a</sup>	0.87
	Water	4.68 <sup>c</sup>	1.01
	Water+cellulase	5.44 <sup>b</sup>	0.85
	Water+lactic acid bacteria	4.35 <sup>g</sup>	0.54
	Water+cellulose+lactic acid bacteria	4.43 <sup>fg</sup>	0.58
SEM		0.03	0.17

Cattle were given 3 kg of silage and measured feed intake as an index of palatability for 4 min.

Means are not significantly different ( $p < 0.05$  Duncan's multiple range test,  $n=3$ ).

shown in Table 4. In both grain and whole crop silages, pH was significantly decreased by supplementation with LA bacteria. Although water soaking treatment also decreased pH value for grain silage, cellulase addition increased pH value for water soaked grain silage. The pH values for whole crop silage with or without any supplements showed approximately 4.5. The pH value for grain silage not soaked in water was the highest of all, and water soaking lowered pH value for grain silage. Palatability of whole crop and grain silages by beef cattle is also shown in Table 4. There were no significant differences in feed intake between any treatment groups.

## DISCUSSION

In the present study, the difference in the quality of fodder rice silage prepared from either whole crop or grain at the same ripening stages was compared. As shown in Table 3, pH value for grain silage at milk-ripe stage was 4.30 and increased by progressing ripening stage. The increase in pH was in good agreement with the findings reported by Nakui et al. (1986a). Table 3 also showed that pH value for whole crop silage was elevated by progressing ripening stage and was lower than that for grain silage especially during latter half of ripening stages. This finding was the first report to compare the difference between grain and whole crop silages at the same ripening stage. As McDonald et al. (1991) reviewed, it has been well known that the amount of moisture present in the ensiled forage crop affects the rate of silage fermentation i.e. the change in pH, concentration of LA, etc). Nakui et al. (1988) also reported that pH value in whole crop silage was strongly influenced by DM content in the material of fodder rice. In general, if the moisture content was reduced to less than 70%, preferably 65%, and if proper exclusion of oxygen was practiced, no additive would be required to lower pH (Horrocks and Vallentine, 1999). According to McDonald et al. (1991), well-preserved unwilted silage was characterized by having low pH values, usually between pH 3.7 and 4.2

and containing high concentration of LA. In the present study, as shown in Table 2, DM content in rice grains was increased by progressing ripening stages, and pH values also increased along with progressing ripening stages. Furthermore, Table 3 showed that changes in LA content in grain silage seemed to be opposite to those in DM contents and pH values. So far, there has been no reports that LA content in grain silage of fodder rice was measured. In this study, therefore, we attempted to determine LA content in grain silage. Statistical analysis indicated that there was significant interaction between silage material (grain and whole crop) and ripening stage on LA content and that only in grain silage LA content was significantly and negatively related to the pH value ( $r = -0.70$ ,  $p = 0.001$ ). This result suggested that changes in pH value in grain silage were resulted by the increase in DM content associated with decreasing LA fermentation.

The original objective in using silage additives was to ensure that the LA bacteria dominated the fermentation, resulting in a well-reserved silage (McDonald et al., 1991). In this respect molasses, which was commercially available, supplied a cheap source of fermentable carbohydrate and was used in our previous research to improve Napiergrass (*Pennisetum purpureum Schum*) silage (Tamada et al., 1999; Yokota et al., 1992, 1998, 2001). Other additives (i.e. sugars such as glucose and sucrose; cereals such as maize, barley, oats, wheat and milo; beet pulp; cell wall degrading enzymes etc.) were also available to supply carbohydrate needed for LA fermentation. When fermentation stimulants such as LA bacteria were added into ensiled forage crop, LA fermentation could be directly stimulated (McDonald et al., 1991). In experiment 2, LA bacteria and cellulase were added into silage to improve the quality of silage. Furthermore, water soaking treatment was also attempted when preparing grain silage because of low DM content in rice grains at full-ripe stage. As shown in Table 4, water soaking treatment could successfully reduce the pH value for grain silage. As McDonald et al. (1991) mentioned that the addition of water to herbage initially stimulated the

growth of bacteria, the treatment might stimulate bacterial growth in rice grain silage as well as grass silage. The pH value in water soaked grain silage supplemented with LA bacteria was significantly lower than that without LA bacteria (Table 4). Therefore, water soaking treatment was supposed to be available to decrease pH value for grain silage prepared from rice grains at full-ripe stage, and supplementation of LA bacteria might enhance the positive effect of water soaking on decreasing pH value.

To evaluate the quality of Napiergrass silage, Manyawu et al. (2003) measured 4 h feed intake as to the index of palatability. In the present study, like Manyawu et al. (2003), palatability of grain and whole crop silages prepared from rice material at full-ripe stage was examined, and there were no significant differences in feed intake between any treatment groups (Table 4). As Colombari et al. (2001) reported that feed intake of low moisture alfalfa silage by lactating cow almost equal to that of high moisture silage, varying moisture did not seem to affect feed intake of rice grain and whole crop silages. Furthermore, although various silage additives could successfully improve silage quality, the correlation equation between silage pH and feed intake was not significant (data not shown). The palatability of fodder rice silage might be related to not only varying pH values but also other factors like LA and VFA contents, and this issue would be elucidated in the future.

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