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Fermentation Quality and *in Vitro* Nutrient Digestibility of Fresh Rice Straw-Based Silage Treated with Lactic Acid Bacteria

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ABSTRACT

The aim of the experiment was to evaluate fermentation characteristics and *in vitro* nutrient digestibility of fresh rice straw-based silage ensiled with addition of epiphytic lactic acid bacteria (LAB) inoculant. The experiment was arranged in a completely randomized design, with 2 × 2 factorial arrangement of treatments. The first factor was the ratio of fresh rice straw (FRS), tofu waste (TW) and cassava waste (CW) consisted of two levels i.e., 40 : 20 : 40 and 40 : 25 : 35, on dry matter (DM) basis). The second factor was the level of LAB inoculant with two levels ie., 0 and 20 mL/kg FM. The treatments were (A) FRS + TW + CW with the ratio of 40 : 20 : 40, without LAB inoculant; (B) FRS + TW + CW with the ratio of 40 : 20 : 40 + LAB inoculant; (C) FRS + TW + CW with the ratio of 40 : 25 : 35, without LAB inoculant; (D) FRS + TW + CW with ratio of 40 : 25 : 35 + LAB inoculant. Results showed that addition of LAB inoculant in silage increased lactic acid concentration (P<0.01) (3.7% vs. 5.2% fresh matter (FM) and V-score (84.6 vs. 89.6) (P<0.05). Concentrations of lactic acid and V-score were higher in silages with high cassava waste (A and B) (P<0.05) compared to silage with low cassava waste (C and D). The IVOMD of rice straw-based silage was affected by the ratio of silage materials and the level of LAB inoculant (P<0.01). There was no interaction between the ratio of silage materials and the level of LAB inoculant (P>0.05) on chemical composition, fermentation quality of silage and *in vitro* digestibility. It was concluded that mixture silage with ratio of 40 : 20 : 40 with the addition of LAB inoculant had the best fermentation quality and nutrient digestibility than other silages.

Key words: digestibility, fermentation, lactic acid, fresh rice straw, silage

ABSTRAK

Tujuan penelitian ini untuk mengevaluasi karakteristik fermentasi dan kecernaan nutrien (*in vitro*) silase berbasis jerami padi segar yang diensilase dengan penambahan bakteri asam laktat (BAL) epifit. Percobaan ini disusun dalam rancangan acak lengkap pola faktorial 2 × 2. Dua faktor perlakuan, yaitu rasio jerami padi segar : ampas tahu : onggok (40 : 20 : 40 dan 40 : 25 : 35, berdasarkan bahan kering (BK)) dan 2 level inokulan BAL (0 dan 20 ml/kg bahan segar). Perlakuan terdiri atas (A) jerami padi segar + ampas tahu + onggok (40 : 20 : 40, berdasarkan bahan kering); (B) jerami padi segar + ampas tahu + onggok (40 : 20 : 40) + inokulan BAL; (C) jerami padi segar + ampas tahu + onggok (40 : 25 : 35); jerami padi segar + ampas tahu + onggok (40 : 25 : 35) + inokulan BAL. Hasil percobaan menunjukkan bahwa penambahan inokulan BAL pada silase meningkatkan konsentrasi asam laktat (3,7% *vs*. 5,2% bahan segar) (P<0,01) dan *V-score* (84,4 *vs*. 89,6) (P<0,05). Konsentrasi asam laktat dan *V-score* lebih tinggi pada silase dengan kadar onggok tinggi (A dan B) (P<0,05) dibandingkan dengan silase dengan kadar onggok rendah (C dan D). Kecernaan bahan organik secara *in vitro* dipengaruhi oleh rasio bahan silase dan level inokulan BAL (P<0,01). Tidak terdapat interaksi antara rasio bahan silase dan level inokulan BAL (P>0,05) pada komposisi kimia, kualitas fermentasi dan kecernaan secara *in vitro*. Disimpulkan bahwa silase dengan rasio 40 : 20 : 40 dengan penambahan inokulan BAL mempunyai kualitas fermentasi dan kecernaan nutrien terbaik dibandingkan silase lain.

Kata kunci: kecernaan, fermentasi, asam laktat, jerami padi segar, silase

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INTRODUCTION

Recently, there is a growing interest in the use of agricultural and food processing industry residues as silage materials. Fresh rice straw (FRS) is the lower part of the rice crop harvested and remained at the rice field. These residues are abundantly available, and most of it are not used as livestock feeds and later are burned. Burned rice straw is then flown to the air and the smoke produced causes air pollution. Li *et al*. (2010) and Zhang *et al*. (2010) reported that rice straw (RS) had potential to be preserved as silage and used as ruminant feeds. However, successful rice straw ensiling is difficult due to low water soluble carbohydrates (WSC) and less epiphytic LAB (Kawamoto *et al*., 2007). Li *et al*. (2010) reported that WSC content of rice straw averaged 19 g/kg of fresh weight. As a general rule, a concentration of 30 g WSC/kg fresh weight is necessary to reduce the risk of secondary fermentation (Chamberlain & Wilkinson, 1996). Sugar-rich materials are commonly used as effective additives for ensiling crops that have low WSC. Cassava waste (CW) is a solid residue produced from cassava powder industry with its primary component of starch. Besides, tofu waste (TW) is a residue resulted from tofu processing and contains high crude protein (CP) on dry matter (DM) basis *i.e.* 21.8±4.5%, and therefore it can be used as a protein source for livestock (Santoso & Hariadi, 2009).

The lactic acid bacteria (LAB) play an important role in silage fermentation and influence silage quality. Under natural circumstances, LAB grows as epiphytic bacteria. However, the population of LAB is usually low and varies with standing crops. Thus, addition of LAB inoculant is needed to improve silage quality (Bureenok *et al*., 2006). In the previous studies, Yahaya *et al*. (2004); Bureenok *et al*. (2006); and Santoso *et al*. (2011a) stated that tropical and temperate forages ensiled with addition of epiphytic LAB inoculant resulted a better fermentation quality as compared to commercial inoculant. Santoso *et al*. (2009) found that lactic acid concentration in grass silage treated with 2% (v/w) of epiphytic LAB inoculant from king grass was higher than silage with addition of LAB inoculant from elephant grass. Similar result was reported in other experiments by Antaribaba *et al*. (2009) and Santoso *et al*. (2011b) that king grass silage and rice crop residue-based silage with addition of epiphytic LAB prepared from king grass had good fermentation quality as compared to control silage. Moreover, Horiguchi & Takahashi (2007) concluded that fermentation quality of green soybean stover silage was improved by addition of 5% (v/w) fermented juice lactic bacteria prepared from green soybean stover. This experiment was carried out to evaluate fermentation characteristic and *in vitro* nutrient digestibility of rice straw-based silage treated with addition of epiphytic LAB inoculant extracted from king grass.

MATERIALS AND METHODS

Inoculant Preparation

The epiphytic LAB inoculant was prepared according to Bureenok *et al*. (2006) as was modified by by Santoso *et al*. (2009); Santoso *et al*. (2011a); and Santoso *et al*. (2012). A 220 g of fresh king grass was macerated in 1000 mL of distilled water by using a high-speed blender for 4 min. The maceration was filtered through two layers of cheesecloths, and 600 mL of filtrate was collected in erlenmeyer containing 18 g of glucose. The filtrate was mixed and incubated anaerobically at 30ºC for 48 h.

Silage Materials and Preparation

Fresh RS (*Oryza sativa* var. Mygongga) was collected from rice field area at Prafi District, Manokwari Regency. The TW and CW were obtained from small-scale food industry located at Prafi District. King grass was harvested at 50 days of regrowth defoliation from the experimental field of Faculty of Animal Science, Fishery and Marine Science, The State University of Papua in Manokwari.

Fresh RS (FRS) was chopped with a domestic cutter into 2-3 cm lengths. The TW and CW were dried by using oven at 60 °C for 48 h before being mixed with fresh FRS. The silages were arranged in a 2 × 2 factorial arrangement of treatments. The first factor was ratio of FRS, TW and CW with two levels i.e., 40 : 20 : 40 and 40 : 25 : 35, on DM basis. The second factor was the level of LAB inoculant consisted of two levels i.e., 0 and 20 mL/kg FM. The treatments were (A) FRS + TW + CW (40 : 20 : 40) without LAB inoculant; (B) FRS + TW + CW (40 : 20 : 40) + LAB inoculant; (C) FRS + TW + CW (40 : 25 : 35) without LAB inoculant; and (D) FRS + TW + CW (40 : 25 : 35)+ LAB inoculant. The FRS, TW and CW ratios used in this study were based on previous study by Santoso *et al*. (2012). The inoculant which contained 10^6 cfu/mL was sprayed onto silage material and subsequently mixed by hand before packing into silos. About 1.5 kg of silage materials were packed into plastic silos (size 295 × 495 × 0.06 mm) and tied with a string. Three replicate silos were prepared for each treatment and stored in room temperature (approximately 28 °C) for 30 d. Samples were collected for preparation of silage extract and sample analyses.

In Vitro Digestibility

Determinations of DM and organic matter (OM) digestibility were conducted by using *in vitro* procedure as previously described by Hariadi & Santoso (2010). Twenty five milliliters of rumen liquor-buffer mixture in a 1 : 4 (v/v) ratio were dispensed in 100 mL glass tubes containing 250 mg of dry sample. Triplicates of blank (with no feed sample) and standard (Pangola grass) were included in each run. Rumen liquor was obtained from two ruminally fistulated Holstein Friesian cross-bred cows fed elephant grass twice a day at maintenance level of DM intake. Rumen liquor was collected in the morning before feeding, mixed and strained through four layers of cheesecloth into a pre-warmed thermos flask. Buffer solution was prepared by dissolving 9.8 g $NaHCO_3$, 9.3 g $NaHPO_4.12H_2O$, 0.47 g NaCl, 0.57 g KCl, 0.04 $CaCl_2$, 0.12 g $MgSO_4.7H_2O$ per 1000 mL distilled water. After gassing CO_2 in the tube, corks were tightly placed over the tubes and were incubated in a water bath at 39°C for 48 h. After 48 h of microbial incubation,

the samples were incubated at 39°C for 48 h with acid-pepsin. Therefore, the contents were filtered through pre-weighed Gooch crucibles and dried at 105°C for 24 h. The percent loss of weight was determined and presented as *in vitro* DM digestibility (IVDMD). The left residue was ashed at 550 °C for determination of *in vitro* OM digestibility (IVOMD).

Chemical Analysis and Microbial Count

Dried samples were used to determine DM, OM and CP according to procedure of AOAC (2005). The fiber content *i.e.*, neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed by using Van Soest *et al*. (1991) method with some modification *i.e.*, NDF was determined without the use of ∞-amylase and sodium sulfite.

The numbers of LAB were counted by pour-plate technique in de Man, Rogosa and Sharpe agar. The plates were incubated at 35°C for 3 d (Bureenok *et al*., 2006).

Preparation of Silage Extract

A 20 g of silage was macerated in 70 mL of distilled water and stored at 4 °C for 24 h. The maceration was then homogenized for 15 min by using a shaker and filtered through a Whatman No. 1542 filter paper. The filtrate was used for determination of pH, volatile fatty acids (VFAs), and lactic acid and NH_3-N. The pH value was determined by using a pH meter (Hanna Hi 9025, Romania). Concentrations of individual VFAs were analyzed by using gas chromatography (Varian CP-9002 GC, Shimadzu, Japan) equipped with flame ionization detector (FID) and stainless steel column (1500 mm × 3 mm i.d). Nitrogen was used as carrier gas at 1.25 kg/cm^2 pressure. The temperatures of injector oven, column oven and detector were 220, 130, and 220 °C, respectively as were previously described by Santoso *et al*. (2012). Concentrations of lactic acid and NH_3-N were analyzed according to colorimetric and Conway methods, respectively as was described by Santoso *et al*. (2011a) and Santoso *et al*. (2012). To assess the quality of the silage, the V-Score was calculated from the NH_3-N/Total N and VFA concentration in the silages (Takahashi *et al*., 2005).

Calculations and Statistical Analysis

Data were analyzed as completely randomized design with a 2 × 2 factorial arrangement of treatments, using GLM procedure of SAS (SAS Institute Inc., Cary, NC). The linear model used for each dependent variable accounted for the effects of silage materials ratio (S), level of LAB inoculant (L), and S × L interaction as fixed effects.

RESULTS AND DISCUSSION

Characteristics of King Grass Extract

The pH value and LAB number in king grass extract before and after 48 h of incubation are shown in Table 1.

Table 1. Changes of pH value and LAB number in king grass extract before and after 48 h of incubation

	Before incubation	After incubation
pH	6.64	4.63
LAB (× 10^6 cfu/mL)	1.60	6.30

The number of LAB in king grass extract after 48 h of incubation increased 4-fold as compared to that before incubation. However, after 48 h of anaerobic incubation, king grass extract had a low pH compared to that before incubation. This result indicated that LAB produced high lactic acid concentration during incubation which resulted in low pH value. This result was consistent with previous studies by Santoso *et al*. (2011a) and Santoso *et al*. (2012) that pH value in king grass extract declined from 6.71 to 3.51 and from 6.81 to 3.29, respectively after incubation at 30°C for 48 h. The declined trend in pH value with the increased LAB number in extracts of grass and legume after 48 h of incubation were also reported by Bureenok *et al*. (2006) and Wang *et al*. (2009).

Chemical Composition of Silages

The nutrients content of rice straw-based silage treated with epiphytic LAB inoculant are presented in Table 2. The moisture content of silages in this study varied from 65.6% to 67.7% which was lower than moisture content of green soybean stover silage with addition of fermented juice of lactic acid bacteria (FJLB) (Horiguchi & Takahashi, 2007). In a study by Takahashi *et al*. (2005), moisture content of whole rice crop silage treated with FJLB was 51.9%. However, addition of dried TW and CW containing low moisture *i.e.*, 13.6% and 13.9%, respectively, may have benefited in increasing DM content of silages from 21.5% to 34%. The DM contents in all silages were higher than the value of 30% for ideal silage as suggested by Chamberlain & Wilkinson (1996). However, the fermentation was also shown to be improved when fresh rice straw was mixed with low moisture CW. Organic matter content of silages increased (P<0.05) with the increased proportion of CW from 35% to 40%. The proportion of fibrous components *i.e.*, NDF and ADF were significantly lower for silage added LAB inoculant (B and D) (P<0.05) than silage without LAB inoculant (A and C), meanwhile hemicellulose content was higher in silages with LAB inoculant addition (P<0.05). One of the explanations for the lower NDF and ADF in those silages was that enzymatic actions *e.g.* hemicellulase and cellulase present in the original forage degraded the cell wall during ensiling (Yahaya *et al*., 2004 and de Oliveira *et al*., 2009). Another reason was the fibrous component that was hydrolyzed and many of the organic acids such as lactic acid and acetic acid, were produced during ensilaging (Cao *et al*., 2010). Similar results were also reported in other experiments using guinea grass, rice straw and king grass silages (Ando *et al*., 2006; Li *et al*., 2010; Santoso *et al*., 2011a). This was also consistent with the results obtained from ensiling of straws of three varieties rice, namely Wuyunjing7, Wuyujing3, and Jinglingxiang

Table 2. Chemical Composition (%) of fresh rice straw and fresh rice straw-based silages

	FRS[1]	Silages[2]				SEM[3]	P-values[4]		
		A	B	C	D		S	L	S×L
Moisture	79.5	67.4	65.6	67.7	67.3	1.26	0.49	0.32	0.62
Organic matter	87.5	90.4	90.9	89.1	89.5	0.54	0.04	0.41	0.93
Crude protein	7.5	9.8	9.8	10.2	10.5	0.49	0.30	0.74	0.79
NDF	77.5	55.7	52.4	53.1	51.9	0.94	0.13	0.04	0.30
ADF	53.0	41.3	37.8	42.9	40.1	1.03	0.10	0.02	0.71
Hemicellulose	7.5	14.4	14.6	10.1	11.8	0.44	<0.01	0.05	0.10

Note:
[1]Fresh rice straw
[2](A) FRS + TW + CW (40 : 20 : 40, on dry matter basis); (B) FRS + TW + CW (40 : 20 : 40) + LAB inoculant; (C) FRS + TW + CW (40 : 25 : 35); (D) FRS + TW + CW (40 : 25 : 35) + LAB inoculant
[3]Standard error of mean
[4]S: effect of silage materials ratio; L: effect of LAB; S×L: interaction between S and L.

treated with LAB inoculant and glucose (Li *et al*., 2010). However, silages with low content of CW (C and D) had lower (P<0.01) hemicellulose content than silages with high content of CW (A and B).

Fermentative Quality of Silages

The fermentation characteristics of FRS-based silage treated with epiphytic LAB inoculant is presented in Table 3. The pH value of silage was affected by the ratio of silage materials (P<0.01) and the level of LAB inoculant (P<0.05). There was no interaction (P>0.05) between the ratio of silage materials and the level of LAB. In our research, pH values in all silages were within the ideal range of 4.0 to 4.5 as was suggested by Chamberlain & Wilkinson (1996). Silage supplemented with LAB inoculant (B and D) had lower pH value (P<0.05) than silage non supplemented with LAB inoculant (A and C). Addition of epiphytic LAB inoculant at ensiling ensured a rapid fermentation which resulted in quick accumulation of lactic acid and low pH at early stages of ensiling. Silage with high content of CW (A and B) had lower pH value (P<0.01) compared to silages C and D. Higher lactic acid in silage with 40% of CW could be attributed to the higher availability of soluble carbohydrate content, thus resulted in high lactic acid concentration produced by LAB. Lower pH value in silages contained 40% of CW or silages treated with LAB inoculant was supported by the higher lactic acid concentrations (P<0.01) in those silages. Danner *et al*. (2003) stated that lactic acid was the strongest of all silage acids and its presence would decrease the pH more effectively than the other VFAs. This result is consistent with previous studies using rice crop residue treated with epiphytic LAB as reported by Takahashi *et al*. (2005) and Santoso *et al*. (2011b). Study reported by Cao *et al*. (2010) showed that the whole crop rice silage added with *L. plantarum* had higher lactic acid concentration than molasses and control silages. McDonald *et al*. (1991) revealed that reducing pH silage prevented the growth of undesirable microbes *e.g*., listeria, clostridia, enterobacteriaceae and moulds that caused a secondary fermentation during ensilage. Sebolai *et al*. (2012) stated that high lactic acid concentration of silage had beneficial for ruminant because *Megasphaera elsdenii* metabolized lactic acid into propionic acid and then was used as a precursor for gluconeogenesis. In addition, LAB inoculant from silage has a potential role as a probiotic which beneficially affects the host animal by improving its intestinal microbial balance (Weinberg *et al*., 2004).

Table 3. Fermentation characteristics of fresh rice straw-based silage after 30 days of ensilage

	Silages[1]				SEM[2]	P-values[3]		
	A	B	C	D		S	L	S×L
pH	4.13	3.97	4.26	4.19	0.04	0.01	0.03	0.11
Organic acids (% FM)								
Lactic acid	4.10	6.70	3.30	3.80	0.55	0.01	0.01	0.10
Acetic acid	0.70	0.63	1.10	0.85	0.12	0.05	0.25	0.50
Propionic acid	0.03	0.03	0.10	0.05	0.03	0.28	0.58	0.51
Butyric acid	0.06	0.04	0.17	0.08	0.03	0.05	0.17	0.36
Total VFAs	0.80	0.73	1.40	0.95	0.12	0.01	0.11	0.16
Total VFAs/Total acids	15.90	10.1	30.40	21.20	3.38	<0.01	0.23	0.13
NH_3-N/Total N (%)	5.80	3.70	5.20	5.10	0.60	0.54	0.19	0.16
V-score	90.70	92.70	78.50	86.00	2.32	0.01	0.05	0.28

Note:
[1](A) FRS + TW + CW (40 : 20 : 40, on dry matter basis); (B) FRS + TW + CW (40 : 20 : 40) + LAB inoculant; (C) FRS + TW + CW (40 : 25 : 35); (D) FRS + TW + CW (40 : 25 : 35) + LAB inoculant
[2]Standard error of mean
[3]S: effect of silage materials ratio; L: effect of LAB; S×L: interaction between S and L.
FM: fresh matter

The acetic acid concentration was lower in silages with 40% of CW (A and B) as compared to silages with 35% of CW (C and D) (P<0.05). This result indicated that the activity of hetero fermentative LAB in both silages A and B was lower than in silages C and D. McDonald *et al*. (1991) revealed that during ensiling, hexose was fermented to lactic acid and other products *i.e*., ethanol and acetic acid. In our research, silages with high content of CW (A and B) had lower butyric acid concentration (P<0.05) than silages C and D. The results indicated that clostridia bacteria were more active in silages with low content of CW than in silages with high content of CW. According to Chamberlain & Wilkinson (1996), secondary fermentation is occurred when insufficient acid is produced by the primary fermentation to reduce the pH to below a critical level of about 4.5. The bacteria responsible for secondary fermentations are mainly the clostridia. These bacteria may convert lactic acid to butyric, or they may degrade proteins, peptides and amino acids to amines and ammonia. McDonald *et al*. (1987) also reported that butyric acid was produced by saccharolytic clostridia *i.e. Clostridium butyricum*. Silage materials with ratio of 40 : 20 : 40 had lower total VFAs concentration as compared to those with ratio of 40 : 35 : 35 (P<0.01). This finding indicated that silage containing FRS, TW and CW in a ratio of 40 : 20 : 40 had better fermentative quality than those in a ratio of 40 : 25 : 35. Chamberlain & Wilkinson (1996) stated that the VFAs consisted of acetic acid, propionic acid, butyric acid 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 amino acid from the skeleton of the amino acid.

In the present study, the proportions of VFAs to total acid were 15.9%, 10.1%, 30.4%, and 21.2%, respectively for silage A, B, C and D. The result indicated that fermentations of silages A and B were better than silages C and D. However, the values found in silages C and D were still above the ideal value of 20% as recommended by Chamberlain & Wilkinson (1996). Percentage of NH_3-N/Total N was not affected by silage materials ration and level of LAB inoculant (P>0.05). However, silage treated with LAB inoculant (B and D) had slightly lower NH_3-N/TN than silage without LAB inoculant (A and C). In the previous work of Bureenok *et al.* (2006) who found that the use of fermented napier grass extract increased lactic acid and greatly inhibited the clostridial activity to protect proteins from extensive degradation. As stated by Givens & Rulquin (2004) that during ensiling, protein was degraded to peptides and free amino acids by plant proteases. In addition, degradation of amino acids to ammonia and non-protein nitrogenous fraction was predominantly due to proteolytic clostridia. Chamberlain & Wilkinson (1996) concluded that ammonia-N was an indicator of the proportion of the total N which was completely degraded during ensiling. Hence, concentration of ammonia-N is the best indicator of secondary fermentation. The growth of proteolytic clostridia, which degrade protein and amino acids to NH_3, is inhibited by low pH (McDonald *et al*., 1991; Givens & Rulquin, 2004). This result was supported by the low pH values in silages B and D compared to silages A and C. This condition could depress the growth of proteolytic clostridia.

The V-score is one of indicator to evaluate the quality of silage. V-scores in silages A and B were significantly higher than those in silages C and D (P<0.01), suggesting that silage with 40% of cassava waste had better fermentative quality as compared to silages with 35% of cassava waste. In addition, V-score found in the present study was comparable with the V-score of whole rice crop silage (Takahashi *et al*., 2005) and slightly lower than total mixed ration silage of whole crop rice (Cao *et al*., 2010).

In Vitro Nutrient Digestibility

Ratio of silage materials and level of epiphytic LAB inoculant had no effect on IVDMD value (P>0.05). On the other hand, IVOMD of rice straw-based silage was affected by the ratio of silage materials and LAB inoculant (P<0.01). However, the IVOMD increased by average of 7.3% when epiphytic LAB inoculant prepared from king grass was added. Increasing IVOMD in silages with addition of LAB inoculant in the present study could be due to the lower NDF and ADF contents. This result was supported by previous study by Ando *et al*. (2006) that addition of LAB increased the digestibility of DM, OM, and CP of guinea grass silage. When compared to rice straw, the IVDMD and IVOMD in rice straw-based silage increased by average of 37.9% and 32.5%, respectively. Silage with high CW content (A and B silages) had higher IVOMD value compared to low CW content (C and D silages) (P<0.01), which may be due to a higher soluble carbohydrate content. In our study, there was no interaction between ratio of silage materials (S) and level of LAB (L).

Table 4. *In vitro* DM and OM digestibility of rice straw and fresh rice straw-based silages

	RS[1]	Silages[2]				SEM[3]	P-value[4]		
		A	B	C	D		S	L	S×L
IVDMD (%)	41.2	56.6	57.4	55.2	58.2	1.20	0.79	0.15	0.40
IVOMD (%)	48.3	65.0	69.9	58.8	62.8	0.75	<0.01	<0.01	0.58

Note:
[1]Rice straw
[2](A) FRS + TW + CW (40 : 20 : 40, on dry matter basis); (B) FRS + TW + CW (40 : 20 : 40) + LAB inoculant; (C) FRS + TW + CW (40 : 25 : 35); (D) FRS + TW + CW (40 : 25 : 35) + LAB inoculant
[3]Standard error of mean
[4]S: effect of silage materials ratio; L: effect of LAB; S×L: interaction between S and L.

CONCLUSION

Addition of LAB inoculant to rice straw-based silage reduced fibrous components *i.e.* NDF and ADF, enhanced lactic acid concentration and OM digestibility (*in vitro*). The mixture silage of FRS, TW, CW in the ratio of 40 : 20 : 40 with addition of LAB inoculant (silage B) resulted a better fermentation quality and *in vitro* digestibility.

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