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EFFECTS OF LACTIC ACID BACTERIA ON ENSILING CHARACTERISTICS, CHEMICAL COMPOSITION AND AEROBIC STABILITY OF KING GRASS

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ABSTRACT

The aim of the present study was to evaluate the effects of lactic acid bacteria (LAB) on ensiling characteristics, chemical composition and aerobic stability of King grass. Silage was prepared in a completely randomized design consisting of threetreatments and one control with three replicates as control (SK, adding 2ml/kg sterilizing water), *Lactobacillus plantarum* commercial bacteria (SKP), *Lactobacillus plantarum* isolated from Napier grass (SKN), *Lactobacillus paraplanetarum* isolated from Italian ryegrass (SKI). All silage were prepared using polyethylene terephthalate bottles, and incubated at room temperature for different ensiling days. The earlier and prolong stage of ensiling, dry matter (DM) was not significantly ($P>0.05$) affected and the level of pH, acetic acid (AA), NH_3-N, water soluble carbohydrate (WSC) and butyric acid (BA) was significantly ($P<0.05$) decreased. Lactic acid (LA), ethanol and propionic acid (PA) was significantly ($P<0.05$) increased in treatments compared to SK. The dry matter (DM), crude protein (CP), propionic acid (PA) neutral detergent fiber (NDF), acid detergent fiber (ADF) did not significantly ($P<0.05$) differ among the treatments at the end of ensiling. Ammonia nitrogen/total nitrogen (NH_3-N/TN) and AA was significantly ($P<0.05$) decreased and LA, and ethanol was significantly ($P<0.05$) increased in the treatments. When the silos were exposure air, the pH was high and LA was numerically lowered but the WSC was not affected. The yeast, mold and LAB were changed significantly ($P<0.05$). It was suggested that adding lactic acid bacteria could improve the fermentation quality of King grass.

Key words: Fermentation qualities, aerobic stability, *Lactobacillus plantarum*.

INTRODUCETION

King Grass (*Pennisetumpurpureophoides*) is one of the tropical grasses of China, characterized by low water soluble carbohydrate, high buffering capacity and poor fermentation characteristics (Yahaya *et al.*, 2004). King grass is a major resource of feed for the dairy animals but the climatic condition normally limiting feed production in Jiangsu province of China. In rainy season, although, the growth rate of King grass is very good, however, in winter its slow growth results in feed shortage (Santoso *et al.*, 2011; Li *et al.*, 2014).

Lactic acid bacteria (LAB) produce lactic acid as a result of water soluble carbohydrate fermentation and play an important role in feed technology by production and preservation of silage. Isolation of wild-type strains from conventional product is a classical method to obtain starter cultures for feed fermentations (Abdelbasset and Djamila, 2008). By using selected wild-type strains, the large-scale production of fermented silage can be developed without losing their unique flavor and particular characteristics (Ammor *et al.*, 2006). Some species of *Lactobacillus* like *Lactobacillus plantarum*, *Pediococcus* species, and *Enterococcus* species. they can improve the level of acidification and fermentation quality by decreasing dry matter loss and protein degradation of grass silages (Driehuis *et al.*, 2001; Wrobel*et al.*, 2008). Aerobic stability is a term that nutritionists have used to define the length of time that silage remains cool and does not spoil after it is exposed to air (oxygen).*Lactobacillus spp.* have been successfully used as an additive to improve the aerobic stability of corn silages (Queiroz *et al.*, 2013). Many researchers have documented the beneficial effect of lactic acid bacteria on the aerobic stability of silages produced from corn (Weinberg *et al.*, 2004), sorghum and barley (Kung and Ranjit, 2001).

The aim of the present study was to evaluate the effects of different strains of LAB inoculation on silage fermentation quality, chemical composition and aerobic stability test of King grass silage.

MATERIALS AND METHODS

Preparation of silage: King grass (*Pennisetumpurpureophoides*) was collected at the middle stage of growth in the experimental grassland of Nanjing Agricultural University China. The grass was chopped in length (1-2cm) with a chopper and ensiled in anaerobic polyethylene terephthalate bottles of 5 litter capacity. Each polyethylene terephthalate bottle contained 3.2 kg of fresh King grass and subjected to the following. Control (SK), *Lactobacillus plantarum* (MTD/1CB, Ecosyl Products Inc. USA commercial

bacteria) (SKP), *Lactobacillus plantarum* isolated from Napier grass (SKN), *Lactobacillus paraplantarum* isolated from Italian ryegrass (SKI). The number of bacteria of each strain was adjusted at $1x10^6$cfu/g.After treating and integration, each treatment (10 samples per treatment) was packed into a polyethylene terephthalate bottles, followed by sealing with a plastic tape and stored at room temperature. Each triplicate silos was opened on day of 1, 3, 5, 7, 14, 30, 60 and 90th.

Chemical Analysis: Dry matter (DM) and crud protein (CP) content of fresh and silage samples were determined by the method ofAOAC (2005) while neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined by the method described by Van Soest *et al.* (1991). Water soluble carbohydrates (WSC) was analyzed by colorimetric after reaction with anthrone reagent (Arthur Thomas, 1977). The pH was calculated using a glass electrode pH meter (pH221, Hanna Ltd., Italian). The contents of lactic acid and NH_3-N were analyzed according to the method of Barker and Summerson (1941) and Chaney and Marbach (1962). The content of VFAs in silage was determined by the method of Shao *et al.* (2005). The silage extracts were centrifuged for 10 min at 10,000 × g, then supernatant was kept for volatile fatty acids (VFAs) analysis, which were determined using gas chromatography equipped with aflame ionization detection (FID) system (Shimadzu GC-17A, with 30 m × 0.25 mm [diameter of film: 0.25_m] capillary column, acid-modified poly [ethylene glycol] phase, GADA-24107, Sigma–Aldrich Co.; conditions: column temperature 125°C, injection temperature 220°C). Buffering capacity of fresh material was calculated by using protocol of Playne and McDonald (1966).

Microbial population: Silage samples (10g) were macerated in 90ml sterilized water using a medium-speed blender for 2 hours. The 100ul macerated extract was added into 900ul serially diluted in sterilized water. Lactic acid bacteria (LAB) were counted on deMan, Rogosa and Sharp (MRS) agar medium (Shanghai Bio-way Technology Co., Ltd.) after incubation in an anaerobic incubator (YQX-II, CIMO Medical Instrument Manufacturing Co., Ltd., Shanghai, China) at 37^0C for 3d. Yeasts were counted on potato dextrose agar (PDA) medium (Shanghai Bio-way Technology Co., Ltd.), and aerobic bacteria were counted on nutrient agar (AN) medium (Qingdao Hope Bio-technology Co., Ltd.) agar plates were kept in incubator at 37^0C for 3d. All microbial data were transformed to log10 and presented on a wet weight basis.

Aerobic Stability Test: Three small silos per treatment were pooled for an aerobic stability test on day 90. Each polyethylene silo was taken at room temperature (12~6^0C), which lasted for 9 days, by the procedure of Ashbell*et al.* (1991). After exposure air, pH, lactic acid (LA), acetic acid, propionic acid (PA) water soluble carbohydrate (WSC), counting of lactic acid bacteria (LAB), aerobic bacteria and yeast production were measured.

Statistical analysis: All statistical analysis was performed using the statistical analysis system (SAS 2003). The statistical significance was set at $P<0.05$. All values were expressed as mean ± standard error of the mean.

RESULTS

Chemical composition of King grass before ensiling and after ensiling is presented in Table 1 and 2 respectively.The pH, acetic acid (AA), water soluble carbohydrates (WSC) and butyric acid (BA) was significantly ($P<0.05$) decreased and lactic acid and propionic acid (PA) was significantly ($P<0.05$) increased in all ensiling days in treatments as compared to control. The chemical compositions and fermentation characteristics of King grass silage is illustrated in Table 3. The dry matter (DM) was not affected among the treatments and control during ensiling. Ethanol is also not affected in ensiling 1,3,5,7 and 14 days in all silages, however, on 30 and 60 days, ethanol was significantly ($P<0.05$) increased in all treatments compared to the control. NH_3-N was significantly ($P<0.05$) decreased in all treatments as compared with control. Water soluble carbohydrates (WSC) was significantly ($P<0.05$) decreasedduring ensiling and in treatments as compared to control. But the commercial LAB was best at day 14, 30 and 60 of ensiling as compared to isolates of Napier and Italian ryegrass lactic acid bacteria. Microbial composition of treatments during ensiling is presented in Table 4. The microbial counting of the King grass silage during different ensiling 1, 3,5,7,14,30 and 60 days had no significant ($P<0.05$) difference between treatment and control.

Chemical composition and fermentation characteristics of King grass on 90 day are shown in Table 5. The dry matter (DM), curd protein (CP), propionic acid (PA) neutral detergent fiber (NDF), acid detergent fiber (ADF) did not differ between the control and treatments. The pH, ammonia nitrogen/ total nitrogen (NH_3-N/total N), WSC and acetic acid (AA) were significantly ($P<0.05$) decreased and lactic acid was significantly ($P<0.05$) increased in the treatmentsas compared to control. Ethanol concentration was significantly ($P<0.05$) increased in all treatments as compared to control. Lactic acid bacteria was significantly ($P<0.05$) increased and yeast and aerobic bacteria was significantly ($P<0.05$) decreased in SKI as compared to SKP, SKN and SK.

The chemical composition of the King grasssilage after aerobic exposure is shown in Figure 1. After 9 days of exposure air, pH showed an increasing

trend in the treatments. The pH in exposure air 6 days before was significantly ($P < 0.05$) lower than that of control, but no significant difference ($P > 0.05$) of the treatments. During exposure air, the content of lactic acid (LA) of the overall downward trend in the treatments (SKP, SKN and SKI) had no significant difference ($P > 0.05$)as compared to control in 9 days ofexposure air. LAB of control was significantly lower than that of SKI ($P < 0.05$) SKN was the lowest. There was no significant ($P < 0.05$) changein the total water soluble carbohydrate content during exposure air, and the water soluble carbohydrate (WSC) content in the control was significantly higher than that SKP, SKN and SKI. But the NH_3-N/TN was increased numerically enhanced as compared to initial value. In figure 2, the aerobic stability characteristics changed significantly ($P<0.05$) after 9 days.

Table 1. Chemical composition of King grass before ensiling.

Items	King grass
DM (g/kg FW)	167.92
CP (g/kg DM)	14.799
NDF (g/kg DM)	808.25
ADF (g/kg DM)	429.81
WSC (g/kg DM)	38.94
Buffering capacity (meq/kg DM)	278.35
LAB (Log cfu/g FM)	5.03
Aerobic bacteria (Log cfu/g FM)	3.68
Yeast (Log cfu/g FM)	4.64

FW, fresh weight; DM, dry matter; FM, fresh matter ; Log, Denary logarithm of the numbers of bacteria.

Table 2.Fermentation qualities of King grassduring ensiling.

Item	Ensiling day	Control (SK)	SKP	SKN	SKI
	1	4.34±0.01a	3.66±0.02c	3.67±0.01c	3.74±0.01b
	3	3.70±0.06a	3.53±0.05b	3.48±0.02b	3.52±0.02b
pH	5	3.67±0.06	3.50±0.04b	3.45±0.02b	3.47±0.04b
	7	3.72±0.04	3.50±0.01	3.54±0.03	3.43±0.03
	14	3.76±0.07a	3.61±0.07b	3.57±0.02b	3.57±0.01b
	30	3.64±0.05a	3.53±0.03b	3.43±0.03bc	3.46±0.03c
	60	3.79±0.05	3.79±0.40	3.52±0.01	3.54±0.05
	1	36.71±0.26c	84.51±2.28a	84.02±9.49a	60.55±5.24b
	3	100.82±6.05b	129.06±13.43a	105.01±3.07b	124.78±11.59a
	5	107.70±15.45b	134.22±9.18a	126.13±4.38ab	112.93±5.49b
Lactic acid g/kg	7	99.20±2.30	99.18±9.77	110.01±11.03	111.67±6.54
	14	100.68±13.80b	117.40±7.49ab	110.68±5.93ab	131.66±16.32a
	30	119.12±10.48	129.54±12.54	139.52±8.24	139±16.82
	60	164.45±31.79	154.68±46.92	162.01±3.34	155.21±31.50
	1	5.76±0.99a	1.62±0.16b	1.53±0.17b	1.02±0.84b
	3	5.56±0.96a	2.81±0.79b	1.88±0.17b	2.87±0.09b
	5	6.78±1.50a	2.75±0.48b	2.61±0.63b	2.62±0.29b
	7	7.42±0.94	2.36±0.64	2.37±0.52	2.87±0.76
Acetic acid g/kg	14	7.18±1.11a	2.45±0.29b	2.25±0.62b	2.90±2.20b
	30	6.91±1.43a	2.78±0.65b	2.59±0.41b	3.42±0.07b
	60	13.30±0.92a	5.90±3.25b	5.23±2.09b	3.22±0.58b
	1	0.56±0.19	1.56±0.12	1.36±0.18	1.12±0.99
	3	050±0.24b	1.36±0.28a	1.52±0.16a	1.17±0.08a
	5	0.35±0.10c	1.34±0.10a	1.41±0.04a	1.06±0.06b
	7	0.31±0.02	0.84±0.15	1.37±0.04	1.89±0.63
Propionic acid g/kg	14	0.37±0.14b	1.39±0.12a	1.57±0.16a	1.63±0.16a
	30	0.43±0.09b	1.96±0.19a	0.70±1.25ab	1.90±0.60a
	60	0.66±0.09a	0.07±0.16b	0.10±0.22b	0.02±0.03b
	1	0.53±0.06a	0.51±0.18ab	0.39±0.07b	0.44±0.29b
	3	0.48±0.42a	0.31±0.02b	0.39±0.16b	0.42±0.03b
	5	0.85±0.44a	0.51±0.04ab	0.44±0.08b	0.41±0.03b
	7	0.84±0.43a	0.67±0.31ab	0.38±0.06c	0.42±0.10b
Butyric acid g/kg	14	0.91±0.02a	0.39±0.08c	0.46±0.07b	0.61±0.45ab
	30	0.76±0.44a	0.51±0.12b	0.57±0.28b	0.54±0.06b
	60	2.14±0.21a	1.55±0.10b	1.37±0.19b	1.43±0.48b

Values with different lower case letters show significant ($p<0.05$). differences among ensiling days in the same treatment.
Group (SK): control (no additive); Group (SKP): *Lactobacillus plantarum: Group* (SKN) (C USA): *Lactobacillus plantarum*; Group (SKI): *Lactobacillus paraplantarum* $1x10^6$cfu/g FW.

Table 3.Chemical compositions and fermentation characteristics of King grass during ensiling.

Item	Ensiling day	Control (SK)	SKP	SKN	SKI
	1	160.63±0.19b	170.39±1.08	190.06±1.53a	170.83±1.02b
	3	170.22±1.26b	180.83±0.40a	180.12±0.67ab	170.66±0.51ab
	5	160.30±2.35	140.87±0.29	160.14±0.71	150.97±0.92
Dry Matter g/kg	7	160.64±0.45	170.59±1.66	170.02±0.60	160.14±1.84
	14	160.54±0.81	160.91±1.03	170.27±0.29	150.55±1.14
	30	150.21±1.29	150.72±0.84	150.46±1.18	140.55±1.13
	60	140.65±1.04	140.83±0.49	150.35±0.86	140.76±0.67
	1	6.66±0.43	5.09±4.79	5.10±1.19	5.95±0.43
	3	7.78±0.74	7.65±3.42	5.60±0.74	8.32±1.12
	5	8.55±2.81	8.23±0.53	8.50±1.87	8.97±2.45
Ethanol g/kg	7	9.03±1.68	10.18±4.43	4.84±0.74	9.18±2.56
	14	8.95±0.37ab	7.94±1.41b	8.99±0.60ab	11.25±2.71ab
	30	6.86±1.70b	10.71±3.04a	11.95±1.67a	10.78±0.25a
	60	11.44±1.96	12.52±6.66	15.25±3.07	12.71±3.12
	1	1.19±0,03a	0.50±0.03b	0.56±0.03b	0.50±0.08b
	3	1.11±0.11a	0.66±0.07b	0.50±0.02b	0.51±0.07b
	5	1.16±0.10a	0.77±0.07b	0.55±0.04b	0.68±0.10ab
NH_3-N/ total N	7	1.37±0.07a	0.91±0.09b	0.89±0.11b	0.87±0.08b
g/kg	14	1.52±0.08a	1.02±0.11ab	0.87±0.12c	1.08±0.07b
	30	1.39±0.18a	1.18±0.16a	0.90±0.04b	1.18±0.10a
	60	1.04±0.18	1.03±0.90	0.37±0.17	0.71±0.19
	1	22.57±1.30	18.67±2.36	18.98±2.32	19.96±2.92
	3	16.66±1.28b	6.15±1.08b	8.25±1.72ab	10.46±0.59a
	5	12.69±0.48b	3.88±0.79b	5.76±1.44a	3.85±0.82b
WSC g/kg	7	11.46±0.33	3.20±1.32	2.16±0.74	2.57±1.85
	14	10.50±1.19b	4.74±1.16a	2.06±1.96ab	2.29±1.21ab
	30	9.11±0.21c	3.46±0.18a	2.35±0.53b	2.46±0.65b
	60	7.61±1.79b	6.13±2.03a	3.04±0.61b	2.24±0.35b

Values with different lower case letters show significant ($p<0.05$).differences among ensiling days in the same treatment.
Group (SK): control (no additive); Group (SKP): *Lactobacillus plantarum: Group* (SKN) (C USA): *Lactobacillus plantarum*; Group (SKI): *Lactobacillus paraplantarum* $1x10^6$cfu/g FW.

Table 4.Microbial compositions of King grassduring ensiling.

Item	Ensiling day	Control (SK)	SKP	SKN	SKI
	1	6.59±0.31b	7.15±0.36a	7.14±0.22	7.00±0.06a
	3	6.72±0.42	6.67±0.67	7.08±0.01	6.52±0.68
	5	6.72±0.66	3.88±0.79	5.76±1.44	3.85±0.82
	7	6.54±0.03a	6.10±0.15ab	5.75±0.50b	5.92±0.34b
LAB log^{10}cfu/g	14	5.71±0.19a	5.20±0.04b	5.22±0.02b	5.26±0.21b
	30	5.15±0.22	5.28±0.26	5.40±0.17	5.00±0.51
	60	5.30±0.24	5.53±0.30	5.00±0.73	4.87±0.19
	1	4.97±0.35	4.30±040	5.04±013	4.74±0.72
	3	3.68±0.14	3.59±0.11	3.99±0.26	3.91±0.53
	5	3.68±0.14	1.15±2.00	1.20±2.00	1.10±1.90
	7	4.94±0.62a	4.00±0.46b	4.66±0.27ab	4.27±0.3ab
Yeast log^{10}cfu/g	14	3.62±0.12	4.07±0.57	3.95±0.56	4.15±.034
	30	4.71±0.10	4.76±0.15	4.56±0.07	5.03±0.51
	60	3.73±0.33	3.92±0.67	3.89±0.19	3.72±0.50
	1	3.75±0.78	3.01±0.24	4.50±0.89	3.83±1.10
	3	2.56±0.22	2.95±0.59	2.78±0.35	3.16±0.40
	5	2.43±0.22	2.69±0.67	2.78±0.35	3.16±0.40
	7	3.83±0.07a	3.37±0.16b	3.74±0.21a	3.74±0.33a
Aerobic bacteria log^{10}cfu/g	14	3.66±0.26	3.23±0.94	2.95±0.88	3.05±0.63
	30	2.62±0.12b	2.59±0.25b	3.47±0.25a	2.90±0.4b1
	60	3.33±0.45	3.67±0.41	3.47±0.18	3.15±0.37

Values with different lower case letters show significant ($p<0.05$).differences among ensiling days in the same treatment. Group (SK): control (no additive); Group (SKP): *Lactobacillus plantarum: Group* (SKN) (C USA): *Lactobacillus plantarum*; Group (SKI): *Lactobacillus paraplantarum* $1x10^6$cfu/g FW.

Table 5.Chemical compositions and fermentation characteristics on 90 days of ensiling.

Items	Control(SK)	SKP	SKN	SKI	Std Error	Sig.
Dry Matter g/kg	160.00	150.20	150.92	140.82	0.527	0.380
pH value	3.87a	3.56b	3.50b	3.47b	0.047	0.001
Lactic acid g/kg	101.05b	132.44a	122.58a	138.28a	7.367	0.032
Acetic acid g/kg	8.09a	3.56b	3.35b	3.89b	0.534	0.001
Propionic acid g/kg	0.61	0.01	0.00	0.03	0.137	0.032
Butyric acid g/kg	1.59a	1.35b	1.21b	1.11b	0.072	0.132
Ethanol g/kg	8.59c	9.18ab	10.87b	11.31a	1.270	0.420
Curd protein (%)	16.05	15.23	15.07	15.40	0.509	0.310
NH_3-N/total Ng/kg	1.62a	1.05ab	0.71b	0.97b	0.179	0.038
WSC g/kg	11.28b	4.36a	3.40a	6.04a	0.930	0.038
NDF g/kg	986.03	987.03	986.81	986.52	0.547	0.616
ADF g/kg	746.42	752.90	721	746.12	27.763	0.864
LAB log^{10}cfu/g	4.66	4.30	4.50	4.49	0.148	0.443
Yeast log^{10}cfu/g	3.37ab	3.56ab	3.56b	4.17a	0.211	0.058
Aerobic bacteria log^{10}cfu/g	3.37a	3.29a	3.03ab	2.46b	0.213	0.122

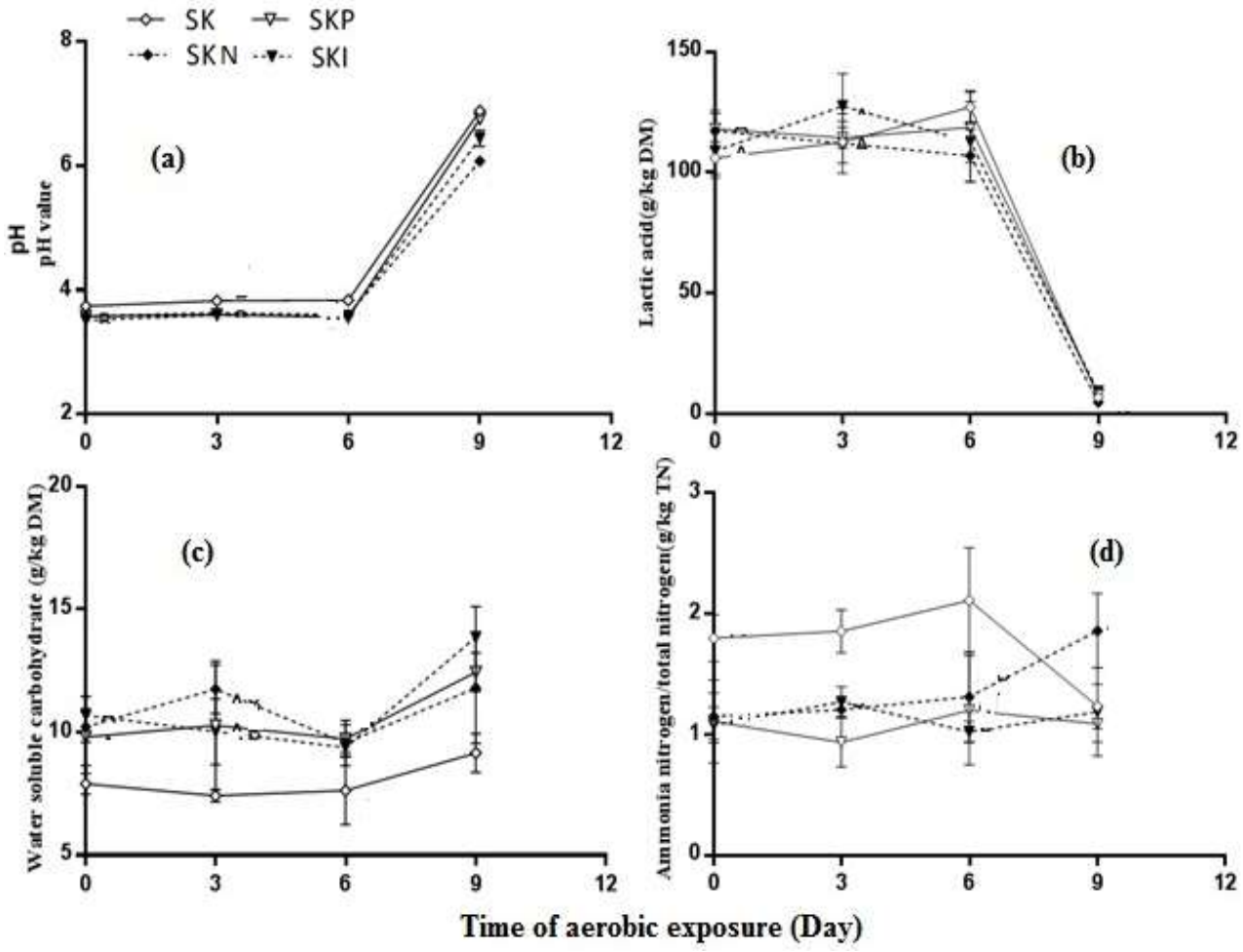

Fig.1 Changes in pH (a), lactic acid (a), water soluble carbohydrate (c) and ammonia/total nitrogen (d) of King grass silage during exposure air.

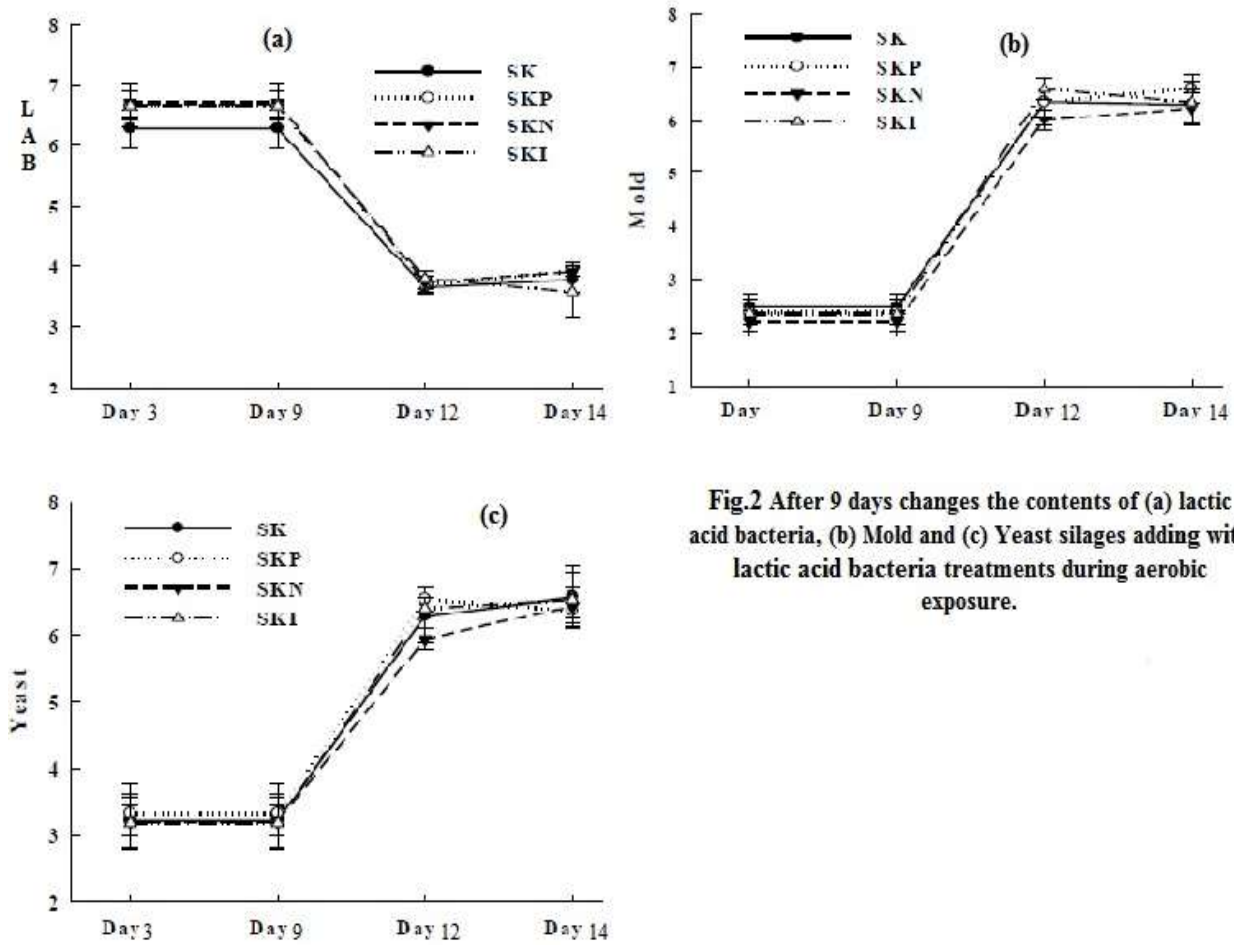

Fig.2 After 9 days changes the contents of (a) lactic acid bacteria, (b) Mold and (c) Yeast silages adding with lactic acid bacteria treatments during aerobic exposure.

DISCUSSION

As observed in this study, the effect of different LAB inoculates in King grass on different ensiling days, pH, acetic acid (AA) and butyric acid (BA) significantly ($P<0.05$) decreased. Similar to our findings, Kim*et al.* (2015) and Yuan*et al.* (2015) reported that the addition of LAB, significantly ($P<0.05$) decreased pH, acetic acid, and butyric acid and the silage DM content was unaffected by bacterial application. Generally, the use of *L. plantarum* is well thought out to be advantageous than the Hetero-fermentative lactic acid bacteria due to its ability to produce a rapid drop in pH and reduction in the NH_3-N concentration (Amanullah*et al.*, 2014).Wang*et al.* (2016) reported lowest pH and highest lactic acid concentration in total mixed ration and whole crop corn silage on day 7, 14, 28 and 56 day of ensiling. Baah *et al.* (2011) reported increased lactic acid concentration and decreased acetic acid, water soluble carbohydrates, butyric acid, NH_3-N and pH in lactic acid bacteria treated.

Homolactic acid bacteria treated silage alone or combination of different strains caused a rapid drop in pH and higher lactic acid concentration (Hassanat *et al.*, 2007; Reich and Kung, 2010). Some researchers also reported that LAB improved the Pearl millet (*Pennisetumamericanum Schum*), Barley *(Hordeum vulgare)* Elephant grass (*Pennisetum purpureum*) and King Grass (*Pennisetum purpureophoides*) silage fermentation (Zahiroddini*et al.*, 2004; Baah*et al.*, 2011).

Zhang*et al.* (2011) observed that a high amount of ethanol accumulation could delay the fermentation quality of the Napier grass silage for the period of the early stage of ensiling. The current study also enhanced the ethanol concentration in the 30 day of ensiling. Filya (2003) found that the number of microbial lactic acid bacteria, yeast and aerobic bacteria numbers were the highest in the *L. plantarum* inoculated silage compared with the control.

Many researchers also observed that the silage DM content was unchanged by inoculation of lactic acid bacteria in prolong period of ensiling (Zahiroddini *et al.*, 2004; Zahiroddini *et al.*, 2006; Baah *et al.*, 2011). Amanullah *et al.* (2014) and Kim *et al.* (2015) reported that during the 100 days of ensiling, there was no significant ($P<0.05$) difference in DM, CP, NDF, ADF. This result of our study agreed with previously reported study where King grass was treated with ELAB and tannin of acacia (Santoso *et al.*, 2011). During the period

of ensiling, protein is degraded to peptides and free amino acid by plant proteases (Owens *et al.*, 2002). In addition, degradation of amino acids to ammonia and non-protein nitrogenous fraction is predominantly due to proteolytic clostridia.

The production of acetic acid (AA), butyric acid (BA) and other acids, is the indication of wasteful fermentation or of secondary fermentation of LA to BA and degradation of amino acid to NH_3 by way of the production of AA from the carbon framework of the amino acid (Chamberlain and Wilkinson, 1996; Santoso *et al.*, 2011). The NDF and ADF were lower in the untreated silage. This result is in concurrence with the previous studies of (Yahaya *et al.*, 2004; de Oliveira *et al.*, 2009; Santoso *et al.*, 2011). One of the justifications for the lower NDF and ADF in the silage is that enzymatic action for example hemicelluloses cellulose present in the original forage on cell wall during ensiling. The concentration of the NDF and ADF was decreased with treated of the LAB had positive effect of silage nutritive value and enhanced digestibility.

The water soluble carbohydrate (WSC) concentration was significantly ($P<0.05$) decreased in all treatments as compared to control after 90 days. Similar results were reported by Adesogan (2006). Generally, increasing WSC concentration can control BA fermentation and promotes the LA fermentation, ultimately improving the silage fermentation quality (Selmer-olsen*et al.*, 1993; Zhang*et al.*, 2010).

The aerobic stability is an important factor in ensuring that silage provides useful nutrients in animal production. When oxygen is exposed to silo, result in spoilage of silage. In the present study, pH showed an increasing trend in the LAB additive after exposure air. Li *et al.* (2016) reported that aerobic weakening occurred within 7 days after exposure air. Hao *et al.* (2015) reported that the weak aerobic stability in additive silage is probably attributable to the high AA and yeast count, which result from the high water soluble carbohydrate concentration. After the exposure to the air, the temperature will enhance in silage and is connected with the microbial oxidation of acid and water soluble carbohydrate to CO_2 and H_2O. In this study, the pH was increased, which is due to the fact that catabolism of protein to ammonia contributes to the enhancement in pH (Swensson, 2003). Liu *et al.* (2015) recently reported that the fermented straw had no effect on the temperature, pH and NH_3-N concentration of the fermented straw. The adsorbed straw had lower pH, AA and high WSC and LA concentration, subsequently it deteriorated much more without difficulty than the fermented straw. Silage that has spoiled because of exposure to air is undesirable due to poorer quality, lower digestibility and the increased risk of the disease and negative effects on the animal production performance. Several studies have been conducted to improve the aerobic stability of silages by inoculating them with *Lactobacillus buchneri* (Holzer *et al.*, 2003; Schmidt *et al.*, 2009; Reich and Kung, 2010).

Conclusion: The inoculation of lactic acid bacteria improved the fermentation quality and aerobic stability of the King grass silage.

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