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The effect of an inoculant and enzymes on fermentation and nutritive value of sorghum straw silages

L. Xing^{a,b}, L.J. Chen^a, L.J. Han^{a,*}

^a College of Engineering, China Agricultural University (East Campus), P.O. Box 191, 17 Qing-Hua-Dong-Lu, Hai-Dian District, Beijing 100083, PR China ^b Chinese Academy of Agricultural Mechanization Sciences, Beijing 100083, PR China

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ABSTRACT

The objectives of this study were to determine the effect of inoculant, enzymes and inoculant-enzymes mixture on fermentation quality, nutritive value, and microbial changes of sorghum straw silage. Sorghum straws were collected and treated with distilled water (control), inoculant, enzymes and inoculant + enzymes prior to ensiling. Three bag silos for each silage (denoted C, I, E and I + E, respectively) were opened after 3, 7, 11, 15, 30 and 60 days for chemical and microbial analyses. For all the silages, there was a rapid decline in pH during the first 3 days of ensiling. Relative to silage C, all the treatment (I, E and I + E) had higher (P < 0.05) lactic acid concentration at all ensiling periods. Population of LAB during all ensiling time was numerically greater for treated than control silages. Separate addition of two additives, especially for enzymes, can effectively (P < 0.05) decrease aNDF and ADF concentration. Treatments with enzymes (E, I + E) can also improve significantly silage IVDMD and IVNDFD concentration, These results indicated that the addition of additives can improve the sorghum straw silage fermentation quality at different extent.

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1. Introduction

In China, there is an annual production of 5 million tons of sorghum straw (MOA, 2006). The sorghum straw, especially for sweet sorghum straw, is an attractive feedstock because as much as 40% of the dry weight consists of the readily fermentable sugars sucrose, glucose and fructose (Henk and Linden, 1992). Therefore, the sorghum straw as feed has an immense potential for China livestock production. For improving the feed quality of sweet sorghum straw, ensiling is one of important methods. Silage additives had been used to improve the silage quality (Cai et al., 1998). Inoculations and enzymes are the most popular silage additives. Previous studies had reported positive outcomes of inoculated silages (Filya, 2003; Guan et al., 2002). Enzymes were originally added to silage to partially degrade fiber to fermentable water soluble carbohydrates (WSC) for use by lactic acid bacteria because these organisms can not use fiber as an energy source to make lactic acid (Eun and Beauchemin, 2007). The mixtures of inoculations and enzymes had also been employed to improve silage fermentation quality (Chen et al., 1994).

Some workers had also reported the effects of additives on the fermentation and nutritive value of whole-crop sorghum silage (Filya, 2003; Guan et al., 2002). In contrast, there is limited study about the effects of inoculant, enzymes and inoculant-enzymes mixture on fermentation and nutritive value of sorghum straw silage. In this study, the straw of sorghum M81E (*Sorghum bicolor*), which is the typical sweet sorghum variety in China, was collected. The series of experiments was undertaken to examine the effect of inoculant, enzymes and inoculant-enzymes mixture on the fermentation and nutritive value of sorghum straw silage.

2. Methods

2.1. Sorghum straw preparation and ensiling

The sweet sorghum M81E (*S. bicolor*) was established in 2003 at the Changping farm of China Agricultural University (longitude 116°23′E, latitude 40°05′N). The sorghum was harvested at the dough stage and threshed. The sorghum straw was chopped to a theoretical length of 20mm using a crop chopper. The chopped sorghum straw was mixed and divided into equal portions for application of four treatments: (1) distilled water (control), denoted as treatment C; (2) inoculant (*Lactobacillus plantarum* Chikuso-1, applied at a rate of 1×10^6 colony forming units (CFU) of lactic acid bacterial (LAB) per gram of fresh material; supplied by Snow Brand Seed Ltd., Sapporo, Japan), treatment I; (3) enzymes (the complex of cellulase and hemicellulase with 90 FPU/ml cellulase activity and 6000 IU/ml xylanase activity, applied at a rate of 0.033 milligram per gram of fresh material which is recommended by



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^{*} Corresponding author. Tel.: +86 10 62736313; fax: +86 10 62736778. *E-mail address*: clj1020@googlemail.com (L.J. Han).

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supplier; supplied by Snow Brand Seed Ltd., Sapporo, Japan), treatment E; (4) inoculant + enzymes, treatment I + E. Treatment I + E was applied in a manner that achieved the same concentrations of inoculant and enzymes as in treatments I and E, respectively. Approximately 100 g portions of sorghum straw from every treatment were packed into plastic film bags (Hiryu KN type, 200 by 300 mm; Asahikasei), and the bags were sealed with a vacuum sealer (BH950, Matsushita). The film bag silos were stored at ambient temperature. Three bag silos per treatment were randomly opened on days 3, 7, 11, 15, 30 and 60 of ensiling to follow fermentation quality.

2.2. Chemical and microbial analysis

Both fresh and ensiled samples (10 g wet basis) were blended with 90 mL of demonized water (20 °C) for 5 min and the pH of water extract was immediately determined using a pH meter (Model HI9024, Hanna Instruments Italia Srl, Italy). Ammonium-N (NH₃-N) concentration was determined as method number 920.03 of AOAC (1990). The colorimetric method was used to determine WSC (Dubois et al., 1956). The concentration of lactic, acetic and butyric acid were measured by high-performance liquid chromatograph method (Ohmomo et al., 1993). The DM content and crude protein (CP, total nitrogen \times 6.25) were determined as method number 934.1 and 990.03 of AOAC (1999), respectively. Neutral detergent fiber (aNDF) were determined according to Mertens method (Mertens, 2002). Acid detergent fiber (ADF) were determined as method number 973.18 of AOAC (1999). In vitro DM digestibility (IVDMD) and NDF digestibility (IVNDFD) were determined using previously described methods (Wilman and Adesogan, 2000). Rumen fluid free of saliva contamination was obtained before morning feeding via rumen fistula from three dry cows. The dry cows were fed a 50:50 alfalfa hat and concentrate diet (DM basis). The diet contained (DM basis) 182 g kg^{-1} CP, 314 g kg⁻¹ aNDF and 36 g kg⁻¹ ether extract. Animals were fed in equal portions at 08:30 and 16:30 h and had free access to water at all times. All chemical analyses were expressed on a dry weight basis, except DM content (g kg⁻¹ fresh matter) and NH₃-N (g kg⁻¹ total nitrogen). The numbers of microorganisms were measured by the plate count method (Cai et al., 1998). Colonies were counted from the plates at appropriate dilutions and the number of colony forming units (CFU) was expressed per gram of fresh forage.

2.3. Statistical analyses

Data of microbial populations were transformed (\log_{10}) but not statistically analyzed because samples were pooled for each forage treatment. The chemical data were subjected to ANOVA by the general linear model procedure of SAS (SAS, 1989). Tukey's test was used to differentiate between means and significance was declared at P < 0.05.

3. Results and discussion

3.1. Chemical composition prior to ensiling

The sorghum straw used for ensiling was characterized by DM content of 235.2 ± 4.9 (g kg⁻¹ fresh matter), concentration of CP of 77.0 ± 2.5 (g kg⁻¹ DM) and concentration of WSC of 156.8 ± 3.6 (g kg⁻¹ DM). The composition of structural carbohydrate in the cell wall was 585.7 ± 5.3 aNDF (g kg⁻¹ DM) and 410.6 ± 4.8 ADF (g kg⁻¹ DM). The relationship between the DM content for silage making and effluent production has been studied by previous workers (Castle and Watson, 1973). The average minimum recommended DM content for ensiling which would produce no effluent or negli-

gible effluent ranges from 229 g kg⁻¹ to 247 g kg⁻¹. In this study, the DM content of the sweet sorghum straw was 235.2 g kg⁻¹ lever which is close to the minimum recommended to ensure successful ensilage with minimal effluent production.

3.2. Lactic acid concentration and pH during the ensiling time

There was a rapid decline in pH for all silages during the first 3 days of ensiling with no significant decline after 3 days post-ensiling for all treatments (Fig. 1). The pH value of all silages after 60 days was below 4. The addition of I, E and I + E at ensiling did not result in a more rapid drop in pH compared to silage C. The lack of response can be attributed to physicochemical properties of sorghum straw. The sweet sorghum straw contained sufficient WSC concentration which can easily produce a rapid drop in silage pH without the use of an additive. The concentration of lactic acid increased rapidly between days 0 and 15 post-ensiling for all silages (Fig. 2). The use of the additives improved significantly lactic acid concentration of silage. Lactic acid bacteria are fast and efficient producers of lactic acid. The addition of enzymes degraded silage cell wall to increase the availability of WSC to serve as a substrate for lactic acid (McDonald et al., 1991).

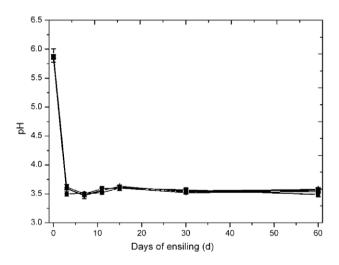


Fig. 1. Changes in pH during ensiling of sorghum straw treated with distilled water $(\mathbf{\nabla})$, inoculant $(\mathbf{\Theta})$, enzymes $(\mathbf{\Box})$ or inoculant + enzymes $(\mathbf{\Delta})$.

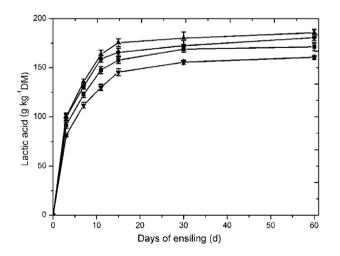


Fig. 2. Changes in lactic acid during ensiling of sorghum straw treated with distilled water (\mathbf{V}) , inoculant $(\mathbf{\bullet})$, enzymes (\mathbf{I}) or inoculant + enzymes (\mathbf{A}) .

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