

IMPROVING UTILIZATION OF SUGAR BEET TOPS AND RICE STRAW BY INOCULATED ENSILAGE

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ABSTRACT

This study was planned to improve the utilization of the underutilized feed resources sugar beet tops (SBT) and rice straw (RS) as feedstuffs for ruminants through processing them into silage. Sugar beet tops and RS were used in different ratios (90,85,80,75% of SBT with 10,15,20,25% of RS) on fresh basis respectively with and without adding silage inoculant. Quality of the prepared silage mixtures was tested at 4, 6, 8 and 12 weeks of ensilage period through determination of pH value and concentrations of lactic, acetic, butyric acids and ammonia-N concentration.

Series of digestibility trials were conducted on 8 bucks (20 kg BW) to determine the TDN value. Rumen fermentation parameters of bucks fed the experimental SBT/RS silage mixtures (pH value, total VFA and ammonia-N concentrations) were evaluated at 0, 3, 6 and 9 h post feeding. Serum metabolites, electrolytes, liver enzyme activities and renal function as affected by feeding the different SBT/RS silage mixtures were determined.

The results of testing silage quality showed that the lowest silage pH value was recorded for inoculated 85% SBT with 15% RS silage mixture. The highest content of lactic acid was recorded for inoculated 85% SBT with 15% RS silage mixture, while the lowest was for uninoculated 75% SBT with 25% RS silage mixture. The lowest content of acetic, butyric acid and ammonia-N was recorded for inoculated 85% SBT with 15% RS silage mixture. The high voluntary DM intake was recorded for bucks fed both inoculated 85% & 90% SBT with 15% & 10% RS silage mixtures. The highest digestion coefficient of DM, CP, NDF, and ADF was recorded for both inoculated 85% & 90% SBT with 15% & 10% RS silage mixtures.

For rumen pH value the lowest was observed for both inoculated 85% & 90% SBT with 15% & 10% RS silage mixtures. Consequently the highest concentration of total VFA was recorded for both inoculated 85% & 90% SBT with 15% & 10% RS silage mixtures, respectively.

Results of the present study indicate that a good quality silage could be obtained from different mixtures of SBT coupled with RS (with and without silage inoculant). Inoculation improves the quality of produced silage, DM intake and TDN values with no adverse effects on rumen fermentation parameters or serum metabolites, serum electrolytes and liver or renal function test.

Key words: Sugar beet tops - Rice straw- Silage quality- Digestibility - Serum metabolites & enzymes

INTRODUCTION

The nutritional status of livestock in Egypt indicates that there is a serious shortage in roughages especially in summer season. To fill this gap all available agricultural by-products should be utilized. Many crop residues are left in the fields to plow under or dumped in the field land and incinerating or drying them is usually not feasible. They always present potential air and water pollution problems. The increased use of these underutilized resources as feedstuffs would keep such problems of disposal and pollution at minimum and add to the cost reduction of rations. Sugar beet tops is one of the underutilized resources, which is produced in large quantities after harvesting the sugar beet crops, especially in Dakahlia and Kafr El-Sheikh governorates. In the season 2000-2001 about 130,000 feddans have been cultivated with sugar beet crops (**Ali and Darwish, 2001**). About 12.5 tons of sugar beet tops are left in each feddan after harvest (**ISBPU, 1990**). Also, a greater attention has been paid to effective use of rice straw as a feed for ruminants, particularly when high quality roughages were too expensive or not available in sufficient amounts. Since the collection and disposal of rice straw is becoming more difficult and expensive, it is left unused as a waste material or simply burned in the fields. Therefore, the use of sugar beet tops with rice straw as a silage may be considered a key solution for some problems of environmental pollution and animal feed shortage.

The main objective of using additives in silage is to obtain lactic acid that rapidly reduce pH to preserve the readily degrading carbohydrates and proteins, inhibit the growth of deteriorating microorganisms (**Van Soest, 1994**) and so produced a good silage with a higher concentration of lactic acid and lower concentrations of acetic, butyric acids and ammonia nitrogen (**Rinne et al., 2002**).

Therefore, the aim of the present study is to evaluate the sugar beet tops (SBT) and rice straw (RS) as feeds for ruminants in form of silage. Silage quality, feed intake and nutritive value of the different SBT/RS silage mixtures were evaluated. The effects of adding silage inoculant (mixture of lactic acid producing bacteria and fibrolytic enzymes) on the nutritional value and fermentation characteristics of the silages were studied. Also, rumen parameters in bucks fed the silage mixtures including pH, total VFA and ammonia-nitrogen concentration were also recorded and studied. Effect of feeding SBT/RS silage mixtures to sheep for 60 days on serum metabolites, electrolytes and enzymes (liver, kidney) were determined.

MATERIALS AND METHODS

Silage mixtures

About two tons of fresh SBT were collected from a sugar beet field at the harvesting time in the mid of April, chopped in pieces of about 5-7 cm length, wilted for two days and used for the preparation of silage with different levels of chopped (2-3 cm) RS. Proximate chemical composition of both SBT and RS are presented in table 1.

Four mixtures of SBT and RS were designed to contain an amount of RS starting with 10% in mixture I and ending with 25% in IV. Each of the four mixtures was divided in to two portions "a" and "b" where LAB-enzymes inoculant was added and mixed with the portion "b". Each of the eight mixtures was tightly packed into double-layer polyethylene bags (50 kg capacity each), tightly closed to maintain the anaerobic condition needed for the proper fermentation. The bags were labeled, piled on a RS bedding and turned down weekly. The mixtures were kept for 3 months.

Determination of silage quality

For judging the quality of the silages and to follow up the course of fermentation, a specified bag from each treatment was used for sampling at 4, 6, 8 and 12 wks of ensilage time. Some of the ensiled mixtures were taken from the center of specific bag for each

mixture, thoroughly mixed for being homogenous and about 1 kg was sampled for testing the silage quality. 100 g of each silage sample was finely homogenized with 1000 ml distilled water in a blender then transferred into a flask of 2 liters capacity provided with a stopper. The homogenized mass was allowed to stand for 20 h with occasional shaking then strained through four layers of cheese cloth. The filtrate was re-filtered through a filter paper several times until it became sufficiently clear to be used for the determination of pH and concentrations of lactic, acetic, butyric acids; and ammonia-nitrogen (**Research Institute for Cattle Feeding at Hoorn Holland, 1961**).

Digestibility trials

Eight apparent healthy mature bucks (20 Kg average body weight) were kept individually in a metabolic stall. Each buck was provided feed and water separately with application of collecting water proof bag for total fecal collection. Two bucks were specified for measuring the digestibility of each silage mixture. Silage was provided *ad lib.* twice daily at 0900 and 1700. Each trial included two sub-periods, preliminary period for 15 days followed by 5 days experimental period in which the daily feed intake and fecal output for each buck were daily recorded at 0900.

Feeding trial

A feeding trial was conducted using sheep (12 weather lambs, averaged 24.5 kg BW) for determination effects of feeding prepared silage (about 2 kg/head) as a part of the daily fattening diet of the lambs (for 8 weeks) on serum metabolites, enzyme activities as indicators of general health, serum electrolytes (Na, K, Cl) were also determined.

Sampling and analysis

Beginning at 0900 of each day of the collecting period, one Kg of silage mixtures was sampled and one tenth of daily fecal output was sampled. Each was mixed, and used for analysis of moisture, CP, EE, CF and ash (**AOAC, 1980**) while NFE and NFC were calculated by difference. Fiber fractions (NDF and ADF) were analyzed (**Goering and Van Soest, 1970**).

Rumen fluid samples were collected at 0 (just before feeding), 3, 6, and 9h post-feeding for 3 successive days at the end of each digestibility trial using stomach tube fitted with 50 ml syringe for suction. The samples were strained through four layers of cheesecloth. Aliquots of

rumen fluid (50ml) were used for determination of pH (Digi-Sense LED pH meter), ammonia-N (**Conway, 1957**), and total volatile fatty acids (**Warner, 1964**) concentrations.

Blood sampling and chemical analysis

1-Blood samples were collected from jugular vein of bucks for 3 successive days before starting the digestibility trials where the bucks fed RS and a concentrate mixture (15% CP, 72% TDN, 0.5% Ca and 0.53% P) and the samples were considered as a control. After 24 h at the end of each digestibility trial blood samples were collected at 4 h post-feeding (for 3 successive days). Blood samples were collected in evacuated centrifuged glass tubes, left for 2 h at room temperature then centrifuge for 10 minutes at 3000 rpm. Sera were carefully aspirated by Pasteur pipette and transferred into dry, clean and sterile labeled glass vials, then kept in a deep freeze until analysis.

2- Biochemical analysis

- a- Serum total proteins (**Henry, 1964**), albumin (**Doumas, 1971**), glucose (**Trinder and Ann, 1969**) were analyzed. Globulin was calculated by difference of total protein and albumin.
- b- Blood samples were collected from sheep (12 weathers) just before feeding the fattening ration including silage mixture as a part of daily ration, then at 30 and 60 days of feeding trial. The blood samples were centrifuged at 3500 rpm for 20 minutes, sera were collected and stored frozen. Serum total lipids (**Zollner and Kirsch, 1962**), cholesterol (**Allain et al., 1974**), triacylglycerol (**Fassati and Precipe, 1982**), urea nitrogen, creatinine and total bilirubin (**Henry, 1974**) were analyzed. The activity of serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) and ALP were determined calorimetrically (**Reitman and Frankel, 1957**). Chemical determinations were carried out using commercial kits (Boehringer, Mannheim Company) and after the procedures described by the instructions of the producer. Sodium, potassium and chloride in the sera samples were also determine using commercial kits (GOD-PAP, Biodiagnostic).

Statistical analysis

All data were statistically analyzed using ANOVA model of SAS software version 6.12 (**SAS, 1996**). Duncan multiple range test was used to test the significance among the means (**Snedecor and Cochran, 1989**).

RESULTS & DISCUSSION

The dry matter contents of the different SBT/RS silage mixtures (Table 3) increased with increasing the level of inclusion of RS at time of ensilage (ranged from 25.6 to 36.38%). **Ohmomo et al. (2002)** reported that ensiled materials should contained 35-40% DM for making good quality silage.

The results in table 4. revealed that there were some losses in DM of the silage in the samples analyzed at time of feeding (15-21 wks of storage) especially for the silage contained high levels of SBT which could be due to effect of aerobic microorganisms, plant and microbial enzymes and fermentation of carbohydrates especially NFC which are of high levels in the high SBT containing mixtures. However, the losses in the inoculated SBT/RS silages were lower than the un-inoculated ones, which might be due to rapid fermentation of soluble carbohydrates with production of lactic acid and rapid decrease in the pH value that inhibited further fermentation, so decreased the more DM losses. **Selmer-Olsen et al. (1993)** reported that inoculation of silage with LAB and fibrolytic enzymes decreased the ensiled DM losses, structural carbohydrates and increased the NFC due to increased the lactic acid production and decreased the silage pH value (**Aksu et al., 2004**).

Regarding the effect of ensilage on carbohydrates, the results in tables 4 showed that the NDF slightly decreased while the NFC increased which could be due to changes in composition of cell wall constituents of ensiled materials due to action of acids produced by fermentation. **Campbell et al. (1990)** detected a considerable increase in WSC during ensilage due to break down of hemicellulose. Addition of LAB inoculants with enzymes to ensiled ingredients decreased the NDF concentration and increased NFC which could be due to effect of LAB and enzymes of the inoculant on the cell wall constituents, which could be hydrolyzed into NFC that promoted the fermentation (Table 4). **Cai et al. (1999)** found that silage inoculant would promote conservation of WSC and increased WSC concentrations.

The CP content of SBT/RS silage mixtures was high in the mixtures contained high levels of SBT and gradually decreased with increasing level of RS which reflected the CP contents of the ensiled materials. The CP contents slightly increased or unaffected by time of storage or inoculant (Table 4). The increase in CP contents in some SBT/RS silage mixtures could be attributed to the losses in DM. EE of different SBT/RS silages were increased due to ensilage (Table 4). The increase in EE contents could be due to the formation of some organic acids and other compounds soluble in organic solvents. Similar findings were

reported by **Bakr (1995)**. The ash contents of the ensiled mixtures were nearly similar which reflected the similar ash contents of the ensiled materials (Tables 2 and 4).

Silage quality:

pH values slightly declined gradually till the end of the 12th wk of ensilage for all mixtures (Table 5) and no further decline was reported in the pH value at time of feeding (15-21 wks of storage). The decrease in the pH values might be due to microbial fermentation of nutrients and formation of acids that inhibit further fermentation. It was reported that addition of the LAB inoculant at ensilage resulted in rapid and vigorous fermentation that resulted in lower pH values and improved forage preservation (**Filya et al., 2000**).

Lactic acid concentration of different SBT/RS silage mixtures (Table 6) revealed that the concentration increased with increasing time of storage till 12th wk, which indicated continuous fermentation and production of lactic acid. Addition of LAB inoculants with enzymes to the ensiled materials enhanced the lactic acid production. Inoculation of the ensiled materials resulted in increased lactic acid concentration which could be due to an increase in NFC by hydrolysis of cell wall, and more rapid decline in the pH value. **Ridla and Uchida (1999)** concluded that the low pH value and high lactic acid in the cellulase added silage.

Acetic acid concentration of the different SBT/RS silage mixtures (Table 7) revealed that the concentration increased with increasing time of storage till 12th wk. Acetic acid concentrations for different SBT/RS silage mixtures were not affected by the level of RS included. The inoculated SBT/RS silage mixtures have lower acetic acid concentration throughout the storage period than un-inoculated ones. **Cai et al. (1999)** observed that when forages were inoculated with LAB before ensilage, the silage usually had a lower content of acetic acid than un-inoculated one.

Concerning the concentrations of butyric acid, the results (Table 7) showed that the butyric acid was undetectable in the SBT/RS silage mixtures till 8th wk of storage. Only very minute concentrations of butyric acid were detected in IVa and IIa SBT/RS silages at 12th wk post-ensilage and their concentrations slightly increased at time of feeding. However, the detected levels of butyric acid were within that of good silage. **Flyn (1981)** stated that good quality silage should have non or traces (< 0.2 to 0.3% of DM) of butyric acid concentration.

The ammonia-N concentrations in the experimental SBT/RS silage mixtures which caused by action of plant and microbial proteases on forage protein (Table 8) indicated low ammonia-N concentrations within the normal levels recommended by **McDonald et al. (1995)** who recorded that ammonia-N for a good quality silage should be less than 2.87% of DM. The changes in SBT/RS silage mixtures due to inoculation showed that LAB inoculated SBT/RS silages with enzymes have lower ammonia-N concentrations than uninoculated ones. The decreased $\text{NH}_3\text{-N}$ concentration in inoculated silages caused by a simultaneous increase in lactic acid production, which decreased pH values and therefore, proteolysis (**Cai and Ohmomo, 1995**).

The DM intake of the different SBT/RS silage mixtures by bucks (Table 9) revealed that the DM intake decreased with increased level of RS in SBT/RS silage mixtures which could be explained by the lower digestibility of RS which resulted in slow disappearance of NDF from the rumen and so physical filling of the rumen. The addition of LAB inoculant to the silage mixtures improved the DM intake especially for high SBT (Iib, Ib and IIIb) silage mixtures. These results could be explained by high lactic acid concentration in inoculated silage mixtures (Table 6). **Bakr (1995)** reported a positive correlation between feed intake and concentration of lactic acids in silages. The results of DM intake (Table 9) revealed that the feed intake from the different silage mixtures was enough to cover the recommended level of DM for maintenance of bucks (2% BW) according to **NRC (1992; 2001)**.

Digestion coefficients of the nutrients of SBT/RS silage mixtures (table 10) revealed that the highest digestion coefficients of DM ($P < 0.05$) were recorded for Iib, Ib and IIIb SBT/RS silages (59.28, 58.31 and 58.12 %, respectively). The digestion coefficient of OM by bucks fed SBT/RS silage mixtures followed the same trend as DM digestibility. **Cai and Ohmomo (1995)** found that the LAB inoculated silage could increase DM and OM digestibilities. The CP digestibility (Table 10) indicated that digestion coefficient decreased with the increased level of RS in SBT/RS silages and the LAB inoculant had no significant effect on digestibility of CP of different silage mixtures.

The digestibility of NDF and ADF of I, II and III SBT/RS silage mixtures were nearly similar and higher than that of IV SBT/RS silages. The results could be explained by the differences in NDF level and composition between the different mixtures (Table 3). Addition of LAB inoculants with enzymes improved the digestibility of NDF and ADF for Ib, Iib and IIIb silage mixture. **Aksu et al. (2004)** found that inoculants stimulated lactic acid production

and formed an appropriate fermentation, consequently the digestibility of NDF and ADF of inoculated silages were higher than that of un-inoculated silages.

The data of TDN values of the silage mixtures (Table 10) revealed that TDN values were higher for Ib and IIb SBT/RS silage mixtures (50.26 and 49.94%, respectively). While, IVa SBT/RS silage had the lowest value (43.54%). The high values of TDN were correlated with high silage quality. **Nefedov (1981)** found that silage quality was correlated with TDN value.

Rumen parameters

Rumen pH values of the bucks fed SBT/RS silage mixtures (Table 11) revealed that the pH value was above 6 at the different sampling times. **Hassanein (1980)** recorded that pH values of ruminal samples collected from goats fed all roughage diets ranged from 6.2 to 7.0. The rumen pH values tended to decrease significantly by advancing the time post-feeding. The lowest values were recorded at 3 h post-feeding. Rumen pH values at 9 h post-feeding returned near to zero h values. **Bakr (1995)** found that the lowest ruminal pH values were at 3 or 6 h post-feeding for rams fed on SBT-silage mixtures and returned to the zero h values by 9 h post-feeding. Feeding of LAB inoculated silages resulted in significant ($P < 0.0001$) lower rumen pH value than un-inoculated ones. This could be due to higher fermentation rates of the inoculated silages. **Weinberg and Muck (1996)** reported that the inoculant treatment decreased rumen pH value and increased total VFA concentrations.

The total VFA in rumen fluid of the bucks fed SBT/RS silage mixtures (Table 12) significantly increased with time post-feeding and the concentration reached the maximum level at 3 h post-feeding then decreased at 9 h post-feeding. It was found that total VFA concentration reached the maximum level at 3-6 h post-feeding of SBT-silages or dried SBT for rams and bucks (**Bakr, 1995 and Orma et al., 1998**).

Feeding the IIb SBT/RS silage resulted in highest total VFA concentration which reflected its high fermentation rate that supported by its lowest ruminal pH value (Table 11). Generally, feeding the LAB inoculated silage mixtures resulted in higher total VFA concentration than uninoculated ones. **Weinberg and Muck (1996)** found that silage inoculant increased total VFA concentration in the rumen liquor.

Ammonia-N concentrations in rumen fluid of the bucks fed the experimental SBT/RS silage mixtures are presented in (Table 13) reached the peak at 3 h post-feeding then gradually

declined at 6 h post-feeding and reached near the values of zero h at 9 h post-feeding. **Bakr (1991)** found that highest concentration of ammonia-N in rumen fluid of bucks fed silage was recorded at 3 h post-feeding. Also, it was reported that the highest concentration of ammonia-N was at 2 h post-feeding in rumen fluid of rams fed corn silage then declined slowly up to 6 h post-feeding (**El-Shinnawy, 2003**). Serum concentrations of metabolites, electrolytes, liver enzymes activities and total bilirubin, creatinine and urea nitrogen of the sheep fed silage mixtures as a part of their daily allowance for 60 days were all within the normal levels. The results showed that feeding of SBT/RS silages had no negative effects on general health of sheep. Similar results were reported by **Bakr (1995)** and **Orma et al. (1998)** in sheep. Generally, results of the present study indicated that a good quality silage could be obtained from different mixtures of SBT coupled with RS (with and without silage inoculant). Inoculated SBT/RS silages showed decreased losses of DM during the storage period and improved the quality of produced silage. It also improved DM intake and TDN values with normal rumen fermentation parameters compared to the uninoculated SBT/RS silages. Inoculation of SBT/RS silage mixtures that contained 85% and 90% SBT with lactic acid producing bacteria and enzymes resulted in improving the quality of produced silage and improved DM intake and TDN values. In addition, there were no adverse effects on rumen fermentation parameters or/ and blood serum metabolites, electrolytes, liver & kidney functions due to feeding these silages. Also, there were no adverse effects of feeding the different silage mixtures on liver and kidney functions (Table 14) in sheep fed the prepared silage mixtures.

Therefore, sugar beet tops coupled with rice straw could be successfully used in preparing silages of considerable quality and good nutritive value especially for mixtures treated with silage inoculant.

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