Research Article

Fermentation Quality and Nutritional Traits of Cluster Bean-Maize Mixture Silages

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Abstract

In present study, effects of different mixture ratios of cluster bean (CB) and maize (M) on silage fermentation and nutritional traits were investigated. Based on dry matter (DM) ratios, maize and cluster bean plants were mixed in five different ratios (100% M, 75% M+25% CB, 50% M+50% CB, 25% M+75% CB and 100% CB). Pure maize, cluster bean and mixture were ensiled for 60 days. Effects of mixture ratios on investigated traits were found to be highly significant (p≤0.01). Increasing silage DM, ADF (acid detergent fiber), NDF (neutral detergent fiber), lactic acid, gas-methane production, metabolizable energy (ME) and organic matter digestibility (OMD) values and decreasing pH levels were observed with increasing maize ratio of the mixtures. Increasing silage crude oil (CO), crude ash (CA) and crude protein (CP) content and volatile fatty acids (acetic, butyric and propionic) were observed with increasing cluster bean ratio of the mixtures. Present findings revealed that improvements were achieved in DM, pH, nutritional traits and ME of the silages with mixtures of maize and cluster bean. It was concluded that 25, 50 and 75% maize ratios in mixtures provided significant contributions to silage quality.

Keywords: Cluster bean, Maize, Silage, Fermentation, Nutritive value

Introduction

Silage offers low loss of nutrients from the harvest to storage, it is easy to use in animal feeding, easy to digest because of fermentation, possible to store for long durations. Thus, silage have become the essential component of lactation...
ration of ruminants to improve milk yields (Mahanna and Chase, 2003). Fermentation is a complex and dynamic process influenced by several factors including anaerobic conditions of the silo, quantity of water-soluble carbohydrates, abundance of epiphytic bacteria, dry matter quantity and buffering capacity of plant material (Weinberg and Muck, 1996). Different methods have been developed for efficient preservation of silage quality with the use of dry matters (Jiang et al. 2020). High lactic acid content of graminae silages results in reduced silage pH, acetic, butyric and propionic acid contents and consequent formation of undesired harmful microbes for fermentation (Mohd-Setapar et al. 2012; Salvucci et al. 2016).

Maize has quite a high biomass yield per unit area, it is easy to harvest, constitute a tasty feed source, thus appetizingly consumed by animals. Maize is highly available for ensilage and maize silage is highly nutritious feed source in ruminant feeding. Therefore, it has become the most important silage crop worldwide (Neylon and Kung, 2003; Kaplan et al. 2016). Although maize silage is an important feed source for dairy cattle, it is poor in protein, thus additives are used to improve protein content of maize silage (Kızılsımsık et al., 2017). Increasing protein content of feed used in rations of high-yield ruminants is a costly practice (Spanghero et al., 2015). Addition of legumes into maize silage increases not only the protein contents, but also the metabolisable energy, taste and the other quality parameters of the silage (Demirel et al., 2009; Blount et al., 2003). Therefore, maize is ensiled in mixtures with soybean (Kızılsımsık et al., 2017), climbing beans (Contreras-Govea et al., 2009), cowpea and bean (Geren et al., 2008), red clover and lupin (Carruthers et al., 2000) and alfalfa (Ozturk et al., 2006).

As it was in the other leguminous crops, cluster bean (Cyamopsis tetragonoloba L.) has also a potential to improve nutritional attributes of maize silage. Cluster bean is quite resistant to high temperatures and droughts (Singh Santosh, 2014). It is rich in pods, leaves and protein, able to be harvested within 3-4 months from sowing and has a high yield potential (Suliman et al., 2017). Cluster bean herbage has a dry matter content of about 26% (Batırca et al., 2017), crude protein ratios of between 16.2-19.8%, ADF ratios of between 38.7-42.9% and NDF ratios of between 43.5-49.8% (Kusvuran et al., 2019). It was reported that cluster bean was not available for grazing because of some harmful substances within the plant structure (Wong and Parmar, 1997). Such a case then necessitates cluster bean to be used as fodder or in silage. However, since cluster bean is a leguminous crop, sole silage has several drawbacks, thus additives should be used to improve quality attributes of cluster bean silage (Oflaz et al., 2019). Although cluster bean is commonly used in animal feeding (Rai, 2015), there aren’t any comprehensive study conducted about mixed silage with maize. Optimal composition of maize and cluster bean mixture silage will provide a great source of feed for livestock, especially for ruminants. In this study, fermentation quality and nutritional attributes of five different mixture ratios of maize and cluster bean silages.

**Materials and methods**

_Forage material and silage and inoculum preparation_

Cluster bean (MA-4246) was harvested at pod set period and maize was harvested at milk-dough stage. Harvested plants were chopped into 2-3 cm long pieces. Chopped samples were homogeneously mixed and placed into 2 kg capacity vacuum bags. Bags were than deaired, closed and kept at dark and room temperature for 60 days. Following 60 days ensilage, pure and mixture silages were analyzed in 3 replicates.

**Chemical and organic acid analyses**

Silage samples (30 g) were mixed with 270 ml distilled water and pH measurements were made. Samples (500 g) were dried in an oven at 70°C for 48 hours. Dried samples were ground to pass 1 mm sieve and made ready for chemical analyses. Crude ash content was determined through ashing samples at 550°C for 8 hours (AOAC 1990). Crude oil content was determined with the use of Soxhlet device. Nitrogen (N) content was measured by the Kjeldhal method. Crude protein ratios of samples were calculated as N × 6.25 (AOAC 1990). Of cell membrane components, ADF and NDF ratios were determined in accordance with the method specified by Van Soest et al. (1991) with the use of ANKOM 200 Fiber Analyzer (ANKOM Technology Corp. Fairport, NY, USA) device.

Acetic, propionic and butyric acid contents were determined with the aid of a gas chromatography device (Shimadzu GC-2010 Kyoto, Japan, column: 30 mm × 0.25 mm × 0.25 μm, Restek, temperature range of 45-230 °C) and lactic acid analyses were conducted with the use of spectrophotometric method (Barker and Summerson, 1941).

**Statistical analysis**

Experimental data were subjected to variance analysis with the use of SAS (1999)
software in accordance with randomized blocks design with 3 replications. Significant means were compared with the use of LSD test.

**Results**

Biochemical characteristics of cluster bean and maize mixture silages are provided in Table 1. Effects of mixture ratios on chemical composition of resultant silages were found to be highly significant (p<0.01). Dry matter decreased, ADF, NDF, CO, CA and CP contents increased with increasing cluster bean ratio of the mixtures (Figure 1). Dry matter ratios varied between 22.02% and 29.17%, ADF contents between 29.88% and 44.12%, NDF contents between 39.36% and 51.20%, CO contents between 1.32% and 3.03%, CA contents between 5.75% and 10.56% and CP contents between 7.79% and 13.03% (Figure 1).

Table 1. Chemical composition of cluster bean and maize mixture silages

<table>
<thead>
<tr>
<th>Mix rates</th>
<th>DM</th>
<th>ADF</th>
<th>NDF</th>
<th>CO</th>
<th>CA</th>
<th>CP</th>
</tr>
</thead>
<tbody>
<tr>
<td>100%CB</td>
<td>22.02a</td>
<td>44.12a</td>
<td>51.20a</td>
<td>3.03a</td>
<td>10.56a</td>
<td>13.30a</td>
</tr>
<tr>
<td>75%CB+25%M</td>
<td>22.34d</td>
<td>42.90b</td>
<td>47.92b</td>
<td>2.72b</td>
<td>8.39b</td>
<td>12.10b</td>
</tr>
<tr>
<td>50%CB+50%M</td>
<td>25.27c</td>
<td>39.12c</td>
<td>44.81c</td>
<td>2.09c</td>
<td>7.76c</td>
<td>11.49c</td>
</tr>
<tr>
<td>25%CB+75%M</td>
<td>28.52b</td>
<td>34.96d</td>
<td>42.52d</td>
<td>1.54d</td>
<td>7.31d</td>
<td>9.37d</td>
</tr>
<tr>
<td>100%M</td>
<td>29.17a</td>
<td>29.88e</td>
<td>39.36e</td>
<td>1.32e</td>
<td>5.75e</td>
<td>7.79e</td>
</tr>
</tbody>
</table>

Sig. ** ** ** ** ** **

LSD 0.48 0.79 0.91 0.05 0.17 0.23

*DM*: dry matter (%); *ADF*: acid detergent fiber (%); *NDF*: neutral detergent fiber (%); *CP*: crude protein (%); *CO*: crude oil (%); *CA*: crude ash (%); *Sig*: significance level; *LSD*: least significant difference; **: p<0.01

![Figure 1. Variation of nutritional traits of cluster bean and maize mixture silages](image-url)
The pH, lactic acid, and volatile fatty acids (VFA) contents of cluster bean and maize mixture silages are provided in Table 2. Effects of mixture ratios on fermentation parameters were also found to be highly significant ($p<0.01$). The pH and VFA contents decreased, and lactic acid contents increased with increasing maize ratio of the mixtures (Figure 1). Silage pH values varied between 3.61% and 5.33%, lactic acid contents between 3.95% and 6.72%, acetic acid contents between 2.48% and 4.54%, propionic acid contents between 0.36% and 0.75%, butyric acid contents between 0.00% and 7.60% (Figure 2).

Biplot graph explained 95.8% total variation in investigated traits. Present traits were placed into 4 groups (BA, pH and AA placed into the same group; DM and LA were placed into the same group; CA, CO and NDF were placed into the same group; CP, PA and ADF were placed into the same group). All traits of mixture silages had equal separation power. While pure maize silage was prominent for LA, DM, pure cluster bean silage was prominent for AA, BA and pH. Mixture silages were positioned in between based on mixture ratios (Figure 3).

Table 2. pH, lactic acid, volatile fatty acids of cluster bean and maize mixture silages

<table>
<thead>
<tr>
<th>Mix rates</th>
<th>pH</th>
<th>LA</th>
<th>AA</th>
<th>PA</th>
<th>BA</th>
</tr>
</thead>
<tbody>
<tr>
<td>100%CB</td>
<td>5.33</td>
<td>3.95</td>
<td>4.54</td>
<td>0.75</td>
<td>7.60</td>
</tr>
<tr>
<td>75%CB+25%M</td>
<td>4.12</td>
<td>4.73</td>
<td>4.28</td>
<td>0.61</td>
<td>2.53</td>
</tr>
<tr>
<td>50%CB+50%M</td>
<td>4.06</td>
<td>4.80</td>
<td>2.80</td>
<td>0.58</td>
<td>0.00</td>
</tr>
<tr>
<td>25%CB+75%M</td>
<td>3.85</td>
<td>4.96</td>
<td>2.61</td>
<td>0.54</td>
<td>0.00</td>
</tr>
<tr>
<td>100%M</td>
<td>3.61</td>
<td>6.72</td>
<td>2.48</td>
<td>0.36</td>
<td>0.00</td>
</tr>
</tbody>
</table>

Sig. Dg.  **  **  **  **  **  **

LSD  0.02  0.10  0.07  0.01  0.08

LA: lactic acid; AA: acetic acid; PA: propionic acid; BA: butyric acid; Sig: significance level; LSD: least significant difference; **: $p<0.01$

Figure 2. Variation of pH, lactic acid, volatile fatty acids of cluster bean and maize mixture silages
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Figure 3. Biplot of variation of nutritional traits of cluster bean and maize mixture silages

Discussion
In present study, effects of different mixture ratios of cluster bean and maize on silage fermentation and nutritional traits were investigated. Even the lowest mixture ratios of cluster bean and maize resulted in sufficient fermentation. In case of mixing two plant species with different dry matter and chemical composition, investigated traits either increase or decrease based on percentages of each silage component (Kizilsimsek et al., 2017). Increasing dry matter, lactic acid, gas-methane production, ME and OMD values and decreasing pH values were observed with increasing maize ratio of the mixtures and increasing ADF, NDF, CA, CO, CP and VFA values were observed with increasing cluster bean ratios of the mixtures.

Ideal DM content of silage materials vary between 25% and 35% (Meyer et al., 1984). In present study, silage DM content were brought to ideal levels with increasing maize ratios of the mixtures. Plant DM content vary with the plant species and largely influenced by plant genetics, cob, leaf and stem ratios, ripening periods, temperature and fertilization programs (Ball et al., 2001; Kaplan et al., 2016). In previous studies conducted with graminae and legume mixture silages, increasing dry matter ratios were reported with increasing graminae ratios of the mixtures (Kizilsimsek et al., 2017; Ozturk et al., 2006; Lima et al., 2010). Besides increasing dry matter ratios, increasing graminae ratios of the mixtures resulted in decreased pH levels (Lima et al., 2010). In present study, pH levels also decreased with lactic acid accumulation. As compared to the other fermentation acids, lactic acid is a desired organic acid and it is the most effective organic acid in reducing pH levels (Umana et al., 1991). Increase in lactic acid production rather than acetic or butyric acid was probably due to increasing ratio of maize with greater water-soluble sugar content than the cluster bean (McDonald et al., 1991). A reverse relationship was reported between pH levels and lactic acid accumulation in quality silages. High lactic acid and low butyric acid ratios are desired in quality silages (McDonald et al., 1991). In present study, lactic acid ratios increased and butyric acid ratios decreased with increasing maize ratios of the mixtures. In quality silages, with sufficient fermentation of lactic acid bacteria, minimum Enterobacteriaceae activity-induced acetic acid production is expected before acidification (Langston et al., 1962). Enterobacteriaceae reduce acetic acid and increase lactic acid concentration (Keshri et al., 2018). Enterobacteria was reported to produce greater quantity of acetic acid in final silage (Kleinschmit and Kung., 2006). In present study, decreasing acetic acid ratios were observed with increasing lactic acid ratios.

NDF and ADF contents largely vary with the type of feed, harvest time and chemical composition (Aderinola et al., 2014). Stalks have greater quantity of cellulose and lignin-like cell membrane components than the leaves and
Leguminosae have greater quantities than graminæ (Aman, 1993). Therefore, increasing ADF and NDF contents were observed with increasing cluster bean ratios of the mixtures (Contreras-Govea et al., 2009). Maize and cluster bean used in present silages have quite different CP, CO, ash, ADF and NDF ratios.

**Conclusions**

It was concluded based on present findings that maize and cluster bean mixture silages were better than pure maize and cluster bean silages. Mixtures improved silage pH, DM, fermentation and nutritional traits. As it was in the other Leguminosae, cluster bean put forth superior nutritional attributes. It was thought that mixture silages could provide significant contributions to animal feeding in intensive livestock operations with high protein needs. Although regardless of mixture ratios, all mixture ratios could be recommended for silage making, it can be recommended that cluster bean plant can be ensiled with maize plant at the ratio of 75%CB+25%M as green herbage basis.

**Conflict of Interest Statement:** The author of the manuscript declares that there is no conflict of interest.

**Contribution Rate Statement Summary:** The writing of the article was performed by the author.

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**References**


