

## A preliminary study on change of mistletoe (*Viscum album* L.) silage quality according to collection time and host tree species

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**Abstract:** In this study, it was aimed to determine the silage quality of mistletoe (*Viscum album* L.) collected from wild pear (*Pyrus amygdaliformis*) and poplar trees (*Populus canadensis*) in January, July, August, and December. The fresh samples were chopped in 2 cm size and ensiled in 2 kg plastic bags then stored at  $25 \pm 2$  °C conditions for 45 days. Silage samples were investigated for dry matter ratio, pH, crude protein ratio, crude ash ratio, condensed tannin, acid detergent fiber, neutral detergent fiber, lactic acid, acetic acid, butyric acid, malic acid, citric acid, succinic acid, potassium, phosphorus, calcium, and magnesium contents. All quality traits of mistletoe silages were significantly different between host trees, collected times, and the interactions of host tree x collection time. It has been determined that mistletoe can be used as silage in terms of all the traits investigated. Besides, the silage of mistletoe collected from wild pear in December is better than other treatments. Mistletoe can be considered as an alternative roughage source due to its high content of crude protein, nutrient composition, easy digestibility, and organic acid content.

**Key words:** Mistletoe, tree species, collection time, silage quality

### 1. Introduction

Increasing animal production all over the world needs more forage day by day but forage production remains under this demand. This situation leads to a quality roughage gap the necessity to use poor quality feeds, which leads to a decrease in animal productivity and quality. The source of these poor-quality roughages may be different, but their common feature is that they have low protein and high fiber content. These constraints point to the search for new resources and the need to evaluate all possible feed sources in rations.

Among the natural forage sources that can be used in animal feeding, leaves obtained from various tree species are important. One of these sources is mistletoe (*Viscum album* L.) for some parts of the world. Mistletoe is a semiparasitic plant that lives on trees and has fodder value with evergreen leaves. It is known the use of mistletoe for animal feeding in regions where traditional farming is the dominant forages source is limited and drought (Madibela, 2009). Mistletoe extracts water and nutrients from the host plant; therefore, it is a rich food source for livestock.

On the other hand, mistletoe contains secondary

metabolites such as flavonoid, phenolic, and condensed tannin. Those are very important for rumen health and animal productivity (Patra et al., 2006; Rochfort et al., 2008; Lee et al., 2017). Robbins (2003) indicated these compounds increased feed intake and animal productivity. Saleh et al. (2015) reported that mistletoe has nutritional and medicinal values for animals. Mistletoe can also be used as silage. Indeed, silage is an important source of feed for livestock when fresh forage is the deficit.

The fermentation should be provided very well to obtain high-quality silage. The oxygen concentration in silo increases decomposition in silage by encouraging fungal activity. Organic acids formed by microorganisms such as beneficial bacteria (lactic, citric, malic, succinic, formic acid, etc.) have the highest growth inhibition efficiency against fungi and yeasts in the silo. Besides, organic acid prevents the silage from spoiling.

The host plant, its photosynthesis and xylem contents are the factors on the diversity of mistletoes (Brodrribb and Holbrook, 2003; Brodrribb et al., 2003). Therefore, this study was aimed to determine the effect of collection time (January, July, August, and December) and tree species (wild pear and poplar) on the silage quality of mistletoe.

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## 2. Material and methods

### 2.1. Material

Mistletoe samples were collected from poplar (*Populus canadensis*) (MFP) and wild pear trees (*Pyrus amygdaliformis*) (MFWP) in January, July, August, and December in the Kurt village of the central district of Bilecik (Turkey) (Figure). Fresh plant materials were chopped in 2 cm size, and samples were compressed in 2 kg plastic bags then stored at  $25 \pm 2$  °C.

#### 2.1.1. Collection of mistletoe

In the determination of the collection times, the periods in which the green forage period decreases or disappears completely, in accordance with the use of the farmers, were taken as a basis. Mistletoe is mostly formed on trees at a height beyond the reach of human hands. For this reason, an auxiliary device was needed to reach the plants and a ladder was used. Plants are harvested with secateurs.

### 2.2. Methods

#### 2.2.1. Dry matter content and pH analysis

After wet weights of silage samples were determined, they were dried in a hot-air oven at 105 °C for 72 h and weighed again for dry matter content (%). The pH of silage samples was determined by using a pH meter. Flieg score ( $\text{Flieg Score} = 220 + (2 \times \text{Dry Matter\%} - 15) - 40 \times \text{pH}$ ) was calculated with a pH and dry matter content (Kilic, 1986). The Flieg score with value for very good was 81–100, for good was 61–80, for the medium was 41–60, for low was 21–40 and for poor was 0–20 represented the silage quality.

#### 2.2.2. Crude protein (CP), acid detergent fiber (ADF), and neutral detergent fiber (NDF) analysis

Silage samples were dried at 65 °C until constant weight. Then, samples were grounded through a hammer mill particle size of about 0.5 to 1 mL. Nitrogen (N) contents of samples were determined using Kjeldahl apparatus (FOSS 984.13) and then multiplying the N concentration by a factor of 6.25 to calculate CP content. The ADF and NDF ratios were determined using ANKOM 200 Fiber Analyzer

device according to Van Soest (1963) and Van Soest and Wine (1967), respectively.

#### 2.2.3. Mineral element analysis

The determination of potassium (K), phosphorus (P), calcium (Ca), and magnesium (Mg) contents were performed by inductively coupled plasma mass spectrometry (ICP-MS) using a Thermo Scientific-iCAPQc (Bremen, Germany).

#### 2.2.4. Lactic, acetic, and butyric acid analysis

The 20 g silage sample was taken from each silage bag and added to 100 mL of distilled water (Başaran et al., 2018). Then silage samples were mixed for 30 min by an electric blender and filtered. Organic acids were analyzed in HPLC (Shimadzu; Kyoto, Japan) autosampler system model LC-20AT equipped with four pumps and an SPDM20A diode array detector (DAD).

#### 2.2.5. Malic, Citric, and Succinic acids analysis

As described by Uden (2018), 100 g of silage sample was refrozen by adding 200 g of water. After dissolving, the liquid part was extracted with a hydrologic press and centrifuged at  $2000 \times g$  for 5 min. Then, supernatants were analyzed in HPLC (Shimadzu; Kyoto, Japan) autosampler system model LC-20AT equipped with four pumps and an SPDM20A diode array detector (DAD).

#### 2.2.6. Total condensed tannin

Tannin solution (6 mL) was added to 0.01 gr of silage sample and mixed on a vortex. The samples were kept in boiling water for 1 h and then at 97–100 °C for 1 h. Finally, they were read at a spectrophotometer at 550 nm (Bate-Smith, 1975). Condensed tannins were calculated by the following formula:  $\text{Absorbance (550 nm} \times 156.5 \times \text{dilution factor) / Dry weight (\%)}$ .

### 2.3. Statistical analysis

The obtained results were subjected to a two-way analysis of variance using the SAS package program (SAS, 1998), and means were separated by Duncan's Multiple Range test.



Figure. The process of collecting mistletoe.

**3. Result and discussion**

Mistletoe samples from poplar (MFP) and from wild pear (MFWP) trees were collected at different times (January, July, August, and December) ensiled at 45 days. The analysis of silage samples showed that host plant, collected time, and host plant x collected time interaction were significant ( $p < 0.01$ ) on the dry matter ratio of silage (Table 1). According to the interaction, the highest dry matter ratio of silage was determined in MFP and MFWP (33.00%) collected in January and in MFP collected in December (32.00%). The dry matter ratio of silage was the lowest in the samples collected in August in MFWP (27.34%) and MFP (26.33%). The dry matter ratio was higher in the winter period compared to the summer period. This is expected because its growth depends on the host tree. Umucalilar et al. (2007) reported that the highest dry matter ratio of mistletoe was collected from 3 different trees (almond, plum, and willow) and 3 different collected times (April, June, and November) was in November. The dry matter of MFWP (29.91%) was lower than that of MFP (30.49%) (Table 1). The good quality silage should contain 25%–40% dry matter. Accordingly, our silages were within the recommended range (Table 1). If the silage contains more than 40% dry matter, palatability decreases with the

high cellulose and hemicellulose content. Besides if the silage contains low dry matter content (<25%), most of the carbohydrates source may be leached (Panyasak and Tumwasorn, 2013).

The pH of mistletoe silages was significantly ( $p < 0.01$ ) affected by the host tree and interaction of host tree x collection time, while collection time was not significant (Table 1). According to the interaction of host tree x collection time, the pH ranged between 4.11–4.84. The average pH value of MFP (4.72) had highest than MFWP (4.37) (Table 1). Filya (2001) reported that the pH value in silage should be below 5 to prevent the proliferation of *Enterobacteria* and *Clostridial* spores, which have a negative effect on fermentation. Aktaş (2012) reported that the pH of the silages that they collected from pear, willow, and wild pear trees were 5.57, 5.32, and 5.22, respectively.

The Flieg score of mistletoe silages was significantly different ( $p < 0.01$ ) between host trees, collected times and, the interactions of host tree x collection time is significant ( $p < 0.05$ ) (Table 2). The highest Flieg score was determined in MFWP collected in December (98.33) and July (97.26), while the lowest was in MFP collected in August (67.00). The average Flieg score of wild pear tree (90.12) was highest than poplar tree (77.13). The Flieg score is determined

**Table 1.** Dry matter ratio and pH of mistletoe silages collected from different trees at different times.

| Months   | Dry matter content (%)** |           |           | pH**     |          |         |
|----------|--------------------------|-----------|-----------|----------|----------|---------|
|          | MFWP                     | MFP       | Average   | MFWP     | MFP      | Average |
| January  | 33.00 a                  | 33.00 a   | 33.00 A** | 4.82 a   | 4.65 a   | 4.74    |
| July     | 30.32 b                  | 30.64 b   | 30.48 B** | 4.21 cd  | 4.62 b   | 4.42    |
| August   | 27.34 d                  | 26.33 d   | 26.84 C** | 4.32 c   | 4.76 ab  | 4.54    |
| December | 29.00 c                  | 32.00 a   | 30.50 B** | 4.11 d   | 4.84 a   | 4.48    |
| Average  | 29.91 B**                | 30.49 A** |           | 4.37 B** | 4.72 A** |         |

\*\* $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

**Table 2.** Flieg score and crude protein content of mistletoe silages collected from different trees at different times.

| Months   | Flieg score** |           |          | Crude protein content (%)** |           |         |
|----------|---------------|-----------|----------|-----------------------------|-----------|---------|
|          | MFWP          | MFP       | Average  | MFWP                        | MFP       | Average |
| January  | 78.06 c       | 85.00 b   | 81.53 B* | 13.14 a                     | 10.62 bc  | 11.88   |
| July     | 97.26 a       | 81.33 bc  | 89.29 A* | 10.61 bc                    | 12.71 ab  | 11.66   |
| August   | 86.86 b       | 67.00 d   | 76.93 B* | 10.65 bc                    | 10.29 bc  | 10.47   |
| December | 98.33 a       | 75.20 cd  | 86.77 A* | 14.16 a                     | 9.15 c    | 11.65   |
| Average  | 90.12 A**     | 77.13 B** |          | 12.14 A**                   | 10.69 B** |         |

\*\* $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

using dry matter content and pH, and gives information on the quality of silage. Kilic (1986) indicated that the Flieg score ranged between 81 and 100 was considered to be very good, between 61 and 80 was considered to be good, between 41 and 60 was considered to be medium, between 21 and 40 was considered to be poor, and between 0 and 20 was considered to be poorer silage quality and excluded from the experiment. Flieg scores of silage determined in this study were found to be a medium, good, and very good quality class of silage (Table 2). Aktaş (2012) reported that the Flieg score of mistletoe silages collected from pear, willow, and wild pear trees ranged from 54.14 to 70.14.

Host plant and interaction of host plant x collection time were significant ( $p < 0.01$ ) on the crude protein content of mistletoe silages, while collection time was not significant (Table 2). Mistletoe silage was determined to be competitive with many quality roughages, with the crude protein content that varied from 9.15% to 14.16%. Yozgatlı et al. (2019) reported that crude protein content of silage maize varieties ranged between 7.09%–9.53%. The average crude protein content of MFWP (12.14%) was higher than MFP (10.69%). This shows that the host tree is an important factor in crude protein in mistletoe. Balabanlı and Karadoğan (1999) reported that the crude protein ratio of mistletoe collected from a pear tree was 14.95%, poplar tree was 13.61%, almond tree was 13.11%, and fir was 8.94%. Madibela et al. (2000) reported that the crude protein ratio of the species of mistletoe ranged between 7.9%–12.8%.

The effects of the host plant ( $p < 0.05$ ) and interaction of host plant x collection time were significant ( $p < 0.01$ ) on the crude ash content of mistletoe silages, while collection time was not significant. The crude ash content was between 9.63% and 10.61% over the collection times and averaged higher in MFWP (10.40%) than MFP (9.94%) (Table 3).

Host tree, collection time, and their interactions were significant ( $p < 0.05$ ,  $p < 0.01$ ) on condensed tannin

content of mistletoe silages (Table 3). High tannin in feed negatively affects protein digestion, microbial, and enzyme activities in ruminants (Kumar and Singh, 1984), however, tannin up to 2%–3% is beneficial to reduce protein degradation in the rumen (Barry, 1987). Önal Aşçı and Acar (2018) indicated that the feeds with low condensed tannin led to increase in protein content of milk, and Li et al. (1996) reported that a low tannin content (0.1%–0.5%) is sufficient to eliminate the risk of rumen swelling after feed consumption. In the present study, the condensed tannin content was below 2% in silages (Table 3). Madibela et al. (2002) reported the condensed tannin contents of *Viscum verrucosum* Harv. and red-berry mistletoe (*Viscum rotundifolium* L.f.) species were 7.5% and 3.1%, respectively. On the other hand, approximately 21%–25% of anthropogenic CH<sub>4</sub> released worldwide is produced in the animal digestive system. Tannins protect proteins from rumen degradation and ruminants excrete less urinary N. Urinary N rapidly converted to ammonia and N<sub>2</sub>O, causing an increase in greenhouse gas emissions, which is an important environmental problem. In this respect, mistletoe silages with a low level of condensed tannins are valuable both as feed and low carbon emission.

The ADF and NDF of mistletoe silages were significantly different ( $p < 0.05$ ) amongst host trees and interactions of host plant x collected time ( $p < 0.01$ ), while collection times were not significant (Table 4). According to the interactions, ADF and NDF ratios ranged between 22.60%–29.82% and 33.88%–40.00%, respectively. The average of ADF and NDF ratios were lower in the silage of MFP (24.38% and 37.95%) compared to the MFWP (26.35% and 38.20%). The best estimate of feed quality relates to the proportion of ADF and NDF in that feed. In other words, it is the amount and ratio of fiber in the feed. The higher fiber in feeds restricts to digest. ADF shows the plant's digestibility, while NDF maturity. In quality feeds, ADF is required to be between 20% and 30% and NDF between 30% and 40% (Cole, 2020). In the present study,

**Table 3.** Crude ash and condensed tannin content of mistletoe silages collected from different trees at different times.

| Months   | Crude ash content (%)** |          |         | Condensed tannin content (%)* |           |           |
|----------|-------------------------|----------|---------|-------------------------------|-----------|-----------|
|          | MFWP                    | MFP      | Average | MFWP                          | MFP       | Average   |
| January  | 9.80 b                  | 11.01 a  | 10.41   | 0.742 a                       | 0.596 bc  | 0.669 A*  |
| July     | 11.52 a                 | 9.71 bc  | 10.61   | 0.645 abc                     | 0.523 c   | 0.584 B*  |
| August   | 9.70 bc                 | 10.42 ab | 10.06   | 0.666 ab                      | 0.569 bc  | 0.617 AB* |
| December | 10.61 ab                | 8.65 c   | 9.63    | 0.628 abc                     | 0.654 ab  | 0.641 AB* |
| Average  | 10.40 A*                | 9.94 B*  |         | 0.670 A**                     | 0.585 B** |           |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

all the silages exhibited ADF and NDF ratios between desired levels (Table 4). Huuskonen et al. (2020) reported that NDF ratio of triticale, barley, and grass silages ranged between 21.3%–55.9%.

The lactic and acetic acid content of mistletoe silages are given in Table 5. According to the host tree x collection time interactions, the highest lactic acid content was determined in MFWP (4.283%) collected in December. When host trees were compared, the silage of MFWP had higher lactic acid (2.310%) than was in MFP (1.838%). The lactic acid content of silages was ranged between 1.055% and 3.398% amongst collection times. The quality of silage highly depends on lactic acid content, and it should be more than 2.0% (Kilic, 2006).

The acetic acid content of mistletoe silages was ranged from 0.028% to 0.156% (Table 5). Alçicek and Özkan (1997) reported that acetic acid indicates the spoiling in silage and its amount should not exceed 0.8%. Aktaş (2012) found that the lactic and acetic acid content of mistletoe silage collected from different host trees (pear, willow, and wild pear trees) were ranged between 1.38%–1.83% and 1.94%–2.03%, respectively. The differences in organic acid contents may be attributed to factors that were subjected to study such as the collection time, host trees, and ecology as well.

Danner et al. (2003) reported that butyric acid is undesirable in the silage because it is the substance with the greatest inhibitory effect on lactic acid bacteria and yeast growth. However, its presence between 0.1% and 0.6% would not affect the silage quality. In the present study, the butyric acid content was ranged between 0.040%–0.205%, and all silages were lower than this critical value (Table 6). Aktaş (2012) reported that the butyric acid content of mistletoe silage collected from pear, willow, and wild pear trees were ranged between 0.89% and 1.26%.

The highest content of malic acid was determined in MFWP collected in December (0.059%) and July (0.049%) and in MFP collected in July (0.047%). Malic acid content was the lowest (0.029%) in MFP collected in January. The malic acid was listed from high to low value according to the collected time: July = December > August > January. Besides, the average malic acid content in the silages of MFWP was higher than was in MFP (Table 6). Diaz-Royon (2012) reported that malic acid improves the ruminal environment and increases propionate production and milk yield of cows. Stallcup (1979) indicated that cows given 70 g/day of malic acid had a higher milk yield. Uden (2018) found that the average malic acid content of maize silage was at 0.05%.

**Table 4.** ADF and NDF ratios of mistletoe silages collected from different trees at different times.

| Months   | Acid detergent fiber (%)** |           |         | Neutral detergent fiber (%)** |          |         |
|----------|----------------------------|-----------|---------|-------------------------------|----------|---------|
|          | MFWP                       | MFP       | Average | MFWP                          | MFP      | Average |
| January  | 26.78 b                    | 24.60 cde | 25.69   | 39.85 ab                      | 38.17 bc | 39.01   |
| July     | 22.60 f                    | 25.65 bcd | 24.13   | 33.88 d                       | 38.32 bc | 36.10   |
| August   | 26.20 bc                   | 23.24 ef  | 24.72   | 39.10 b                       | 37.21 c  | 38.15   |
| December | 29.82 a                    | 24.06 def | 26.94   | 40.00 a                       | 38.08 bc | 39.04   |
| Average  | 26.35 A*                   | 24.38 B*  |         | 38.20 A*                      | 37.95B*  |         |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

**Table 5.** The lactic and acetic acid content of mistletoe silages collected from different trees at different times.

| Months   | Lactic acid (%)** |           |           | Acetic acid (%)** |          |           |
|----------|-------------------|-----------|-----------|-------------------|----------|-----------|
|          | MFWP              | MFP       | Average   | MFWP              | MFP      | Average   |
| January  | 2.493 b           | 2.968 b   | 2.731 B** | 0.031 c           | 0.035 c  | 0.033 C** |
| July     | 1.270 bc          | 0.960 c   | 1.115 C** | 0.130 ab          | 0.156 a  | 0.143 A** |
| August   | 1.197 bc          | 0.913 c   | 1.055 C** | 0.131 ab          | 0.115 b  | 0.123 B** |
| December | 4.283 a           | 2.512 b   | 3.398 A** | 0.028 c           | 0.039 c  | 0.033 C** |
| Average  | 2.310 A**         | 1.838 B** |           | 0.080 B*          | 0.084 A* |           |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

The citric and succinic acid content of mistletoe silages were significantly ( $p < 0.05$ ) affected by host tree, collection time and interaction of host tree x collection time was important (Table 7). Over the interactions, the citric acid of mistletoe silages was ranged from 0.204% to 0.286% and averaged higher in MFWP than MFP. Citric acid contains active ingredients with antimitotic activity for livestock and ensures that the pH remains between 4 and 6 during the fermentation of the silage (Kung et al., 1998; Uden, 2018). Playne and McDonald (1966) found that citric acid of Italian ryegrass silage between 0.1% –2.5%.

The succinic acid of mistletoe silages was ranged between 0.021%–0.029%. It was listed from high to low value according to the collected time: July = December > January = August. The average content of succinic acid in silage of MFWP (0.025%) was higher than MFP (0.024%) (Table 7). Succinic acid is a well-known agent for silage fermentation (McDonald et al., 1991) and is effective for the various diseases of the livestock, and it contributes to the development of the body growth of livestock. Uden (2018) reported that the succinic acid of legumes and silage maize ranged between 0.01% and 0.09%.

The effect of the host tree and collection time and their interaction on K and P contents of mistletoe silage was

significant (Table 8) ( $p < 0.01$ ,  $p < 0.05$ ). The K content was noted the highest (3.222%) in the MFWP collected in July, and the lowest (2.041%) in the MFP collected in December. The P content of mistletoe silages ranged between 0.374% and 0.552% in terms of interaction. Over the collection time, the average K and P of MFWP (2.637% and 0.474%, respectively) were higher than of MFP (2.437% and 0.441%) (Table 8). Ahemad et al. (2009) and Yogeshpriya and Selvara (2018) reported that P is involved in every metabolic reaction and energy transfer within the animal body, while K plays an important role in osmotic pressure regulation and water balance in the animal's body. Accordingly, the roughage requires at least 0.21% of P and 0.8% of K (Kidambi et al., 1993; Tekeli and Ateş, 2005). In the current study, the K and P content of all silages was at the desired level (Table 8).

The Ca and Mg contents of mistletoe silages are shown in Table 9. According to the interaction, Ca and Mg contents were ranged between 0.782%–1.072% and 0.216%–0.347%, respectively. The MFWP was more than MFP in terms of both mineral nutrients. Kidambi et al. (1993) and Tekeli and Ateş (2005) reported that roughage requires at least 0.3% of Ca and 0.1% of Mg.

**Table 6.** Butyric and malic acid content of mistletoe silages collected from different trees at different times.

| Months   | Butyric acid* |           |          | Malic acid** |           |          |
|----------|---------------|-----------|----------|--------------|-----------|----------|
|          | MFWP          | MFP       | Average  | MFWP         | MFP       | Average  |
| January  | 0.040 e       | 0.050 e   | 0.045 C* | 0.034 de     | 0.029 e   | 0.032 C* |
| July     | 0.096 cd      | 0.205 a   | 0.151 A* | 0.049 ab     | 0.047 abc | 0.048 A* |
| August   | 0.056 de      | 0.123 c   | 0.112 B* | 0.036 cde    | 0.045 bcd | 0.041 B* |
| December | 0.133 bc      | 0.167 ab  | 0.150 A* | 0.059 a      | 0.037 cde | 0.048 A* |
| Average  | 0.813 B**     | 1.363 A** |          | 0.045 A**    | 0.039 B** |          |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

**Table 7.** Citric and succinic acid content of mistletoe silages collected from different trees at different times.

| Months   | Citric acid* |          |           | Succinic acid* |           |          |
|----------|--------------|----------|-----------|----------------|-----------|----------|
|          | MFWP         | MFP      | Average   | MFWP           | MFP       | Average  |
| January  | 0.219 bc     | 0.271 ab | 0.245 AB* | 0.022 bc       | 0.026 abc | 0.024 B* |
| July     | 0.273 ab     | 0.225 bc | 0.249 A*  | 0.027 ab       | 0.022 bc  | 0.025 A* |
| August   | 0.204 c      | 0.218 bc | 0.211 B*  | 0.021 c        | 0.022 bc  | 0.022 B* |
| December | 0.286 a      | 0.267 ab | 0.277 A*  | 0.029 a        | 0.027 ab  | 0.028 A* |
| Average  | 0.246 A*     | 0.245 B* |           | 0.025 A*       | 0.024 B*  |          |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

**Table 8.** Potassium and phosphorus content (%) of mistletoe silages collected from different trees at different times.

| Months   | Potassium** |           |           | Phosphorus** |           |          |
|----------|-------------|-----------|-----------|--------------|-----------|----------|
|          | MFWP        | MFP       | Average   | MFWP         | MFP       | Average  |
| January  | 2.312 cd    | 2.597 bc  | 2.454 BC* | 0.424 c      | 0.476 b   | 0.450 B* |
| July     | 3.222 a     | 2.376 cd  | 2.799 A*  | 0.552 a      | 0.456 bc  | 0.504 A* |
| August   | 2.507 bcd   | 2.717 b   | 2.612 B*  | 0.460 bc     | 0.458 bc  | 0.459 B* |
| December | 2.507 bcd   | 2.041 d   | 2.274 C*  | 0.459 bc     | 0.374 d   | 0.417 C* |
| Average  | 2.637 A**   | 2.432 B** |           | 0.474 A**    | 0.441 B** |          |

\*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . There is not a difference between same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

**Table 9.** Calcium and magnesium content (%) of mistletoe silages collected from different trees at different times.

| Months   | Calcium** |           |         | Magnesium** |           |         |
|----------|-----------|-----------|---------|-------------|-----------|---------|
|          | MFWP      | MFP       | Average | MFWP        | MFP       | Average |
| January  | 0.886 bcd | 0.995 ab  | 0.941   | 0.245 de    | 0.275 c   | 0.260   |
| July     | 0.899 bcd | 0.958 b   | 0.929   | 0.298 b     | 0.265 cd  | 0.282   |
| August   | 0.911 bc  | 0.810 cd  | 0.861   | 0.265 cd    | 0.226 ef  | 0.246   |
| December | 1.072 a   | 0.782 d   | 0.927   | 0.347 a     | 0.216 f   | 0.282   |
| Average  | 0.942 A** | 0.886 B** |         | 0.288 A**   | 0.246 B** |         |

\*\* :  $p < 0.01$ . There is not a difference between the same letters in each column ( $p < 0.05$ ). MFWP: Mistletoe from wild pear; MFP: Mistletoe from poplar.

#### 4. Conclusion

From the above study, it was concluded that mistletoe can be considered as an alternative roughage source due to its high content of crude protein, nutrient composition, easy digestibility, and organic acid content. Besides, it has been determined that the silage of mistletoe from wild pear trees in December is better than other treatments.

On the other hand, it was observed that the density of mistletoe in host plants during the collection study was different between regions. This may be due to ecological differences. In addition, birds are an important factor in the spread of mistletoe. For this reason, the bird population in the region may also be effective in the mistletoe density. As

a result, mistletoe silage is a source of quality roughage that can be used successfully in animal nutrition, depending on the harvest time and the host plant. However, its use at an economic level may be valid for certain areas, as its presence varies depending on the region and host plant density.

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#### References

- Ahemad M, Zaidi A, Saghir Khan M, Oves M (2009). Biological importance of phosphorus and phosphate solubilizing microbes - an overview. In: Khan MS, Zaidi A. (editors). Phosphate Solubilising Microbes for Crop Improvement. Newyork, USA: Nova Science Publishers, pp. 1-14.
- Aktaş F (2012). Determination of Feed Values and Energy with In-Situ and In-Vitro Techniques, Silage Quality of Common Mistletoe and Its Silage. Msc, Ondokuz Mayıs University, Samsun, Turkey.
- Alçicek A, Özkan K (1997). Determination of Silage Quality for Silo Feed. In "First Turkish Sillage Conference" pp. 241-247.
- Balabanlı C, Karadoğan T (1999). Possibilities of using mistletoe *Viscum album* L as animal feed. Selcuk Journal of Agriculture and Food Sciences 13: 101-106.
- Barry TN (1987). Secondary compounds of forages. In: Hacker JB, Ternouth JH. (editors). Nutrition of Herbivores. Sydney, Australia: Academic Press, pp. 91-120.

- Brodrribb TJ, Holbrook NM (2003). Stomatal closure during leaf dehydration, correlation with other leaf physiological traits. *Plant Physiology* 132: 2166-2173.
- Brodrribb TJ, Holbrook NM, Edwards EJ, Gutierrez MV (2003). Relations between stomatal closure, leaf turgor and xylem vulnerability in eight tropical dry forest trees. *Plant Cell Environ* 26: 443-450.
- Başaran U, Gülümser E, Mut H, Çopur Doğrusöz M (2018). Determination of Silage Yield and Quality of Grasspea + Cereal Intercrops. *Turkish Journal of Agriculture - Food Science and Technology* 6 (9): 1237-1242.
- Bate-Smith EC (1975). Phytochemistry of proanthocyanidins. *Phytochemistry* 14: 1107-1113.
- Cole E (2020). Understanding Your Forage Test. Available at: <http://extension.missouri.edu/webster/documents/resources/agriculture/UnderstandingYourForageTest.pdf>. [verified 13 April 2020].
- Danner H, Holzer M, Mayrhuber E, Braun R (2003). Acetic acid increases stability of silage under aerobic conditions. *Applied and Environmental Microbiology* 69: 562-567.
- Diaz-Royon F (2012). Effect of malic acid in dairy cow diets. *All About Feed* 24 (10): 6-7.
- Filya I (2001). Silage technology. Hakan Ofset, İzmir, Turkey.
- Huuskonen A, Jaakkola, S, Mann K (2020). Intake, gain and carcass traits of Hereford and Charolais bulls offered diets based on triticale, barley and grass silages. *Agricultural and Food Science* 29: 318-330.
- Kidambi SP, Matches AG, Karnezos TP, Keeling JW (1993). Mineral concentrations in forage sorghum grown under two harvest management systems. *Agronomy Journal* 85: 826-833.
- Kilic A (1986). Silo feed (Instruction, Education and Application Proposals). Turkey, İzmir Bilgehan Press.
- Kilic A (2006). Determined of quality in roughage. İstanbul, Turkey, Hasad Publication.
- Kumar R, Singh M (1984). Tannins: Their adverse role in ruminant nutrition. *Journal of Agricultural and Food Chemistry* 32: 447-453.
- Kung JR, Sheperd AC, Smagala AM, Enders KM, Bessett CA et al. (1998). The effect of preservatives based on propionic acid on the fermentation and aerobic stability of corn silage and a total mixed ration. *Journal of Dairy Science* 81: 1322-1330.
- Lee SHY, Humphries DJ, Cockman DA, Givens DI, Spencer JPE (2017). Accumulation of citrus flavanones in bovine milk following citrus pulp incorporation into the diet of dairy cows. *EC Nutrition* 7 (4): 143-154.
- Li YG, Tanner G, Larkin P (1996). The DMACA-HCl protocol and the threshold proanthocyanidin content for bloat safety in forage legumes. *Journal of the Science of Food and Agriculture* 70: 89-101.
- Madibela OR, Boitumelo WS, Letso M (2000). Chemical composition and in vitro dry matter digestibility of four parasitic plants in Botswana. *Animal Feed Science and Technology* 84: 97-106.
- Madibela OR, Letso M, Boitumelo WS, Masedi M, Alton K (2002). Chemical composition of four parasitic plants harvested over a period of 6 months from two sites in Botswana. *Animal Feed Science and Technology* 95: 159-167.
- Madibela O (2009). Chemical composition and in vitro dry matter digestibility of four parasitic plants (*Tapinanthus lugardii*, *Erianthenum ngamicum*, *Viscum rotundifolium*, *Viscum verrucosum*) in Botswana. *Animal Feed Science and Technology* 84 (1): 97-106.
- McDonald P, Henderson AR, Heron SJE (1991). *Biochemistry of silage*. Marlow: Chalcombe Publication pp. 1991-340.
- Önal Aşçı Ö, Acar Z (2018). Inorganic substances found in the structure of the plant. In: Önal Aşçı Ö, Acar Z. (editors). *Minerals*. Ankara, Turkey: Agriculture engineers chamber press, pp. 68-69.
- Panyasak A, Tumwasorn S (2013). Effect of Moisture Content and Storage Time on Sweet. *Walailak Journal of Science and Technology* 12 (3): 237-243.
- Patra AK, Kamra DN, Agarwal N (2006). Effect of plant extracts on in vitro methanogenesis, enzyme activities and fermentation of feed in rumen liquor of buffalo. *Animal Feed Science and Technology* 128 (3-4): 276-291.
- Playne MJ, McDonald P (1966). The buffering constituents of herbage and of silage. *Journal of Science of Food and Agriculture* 17: 264-268.
- Robbins RJ (2003). Phenolic acids in foods. *Journal of Agricultural and Food Chemistry* 51: 2866-2887.
- Rochfort S, Parker AJ, Dunshea FR (2008). Plant bioactives for ruminant health and productivity. *Phytochemistry* 69 (2): 299-322.
- Saleh I, Maigandi SA, Hudu MI, Abubakar MI, Shehu AU (2015). Uses and chemical composition of mistletoe (*Viscum album*) obtained from different species of trees. *Dutse Journal of Agriculture and Food Security* 2 (1): 8-12.
- SAS Institute (1998). SAS proprietary software. Release 8.2. SAS Institute. Cary, North Carolina, USA.
- Stallcup OT (1979). Influence of addition of dl-malic acid to diets of lactating dairy cows. *Journal of Dairy Science* 62 (Suppl. 1): 225-226.
- Tekeli AS, Ateş S (2005). Yield potential and mineral composition of white clover (*Trifolium repens* L.) – tall fescue (*Festuca arundinacea* Schreb.) mixtures. *Journal of Central European Agriculture* 6: 27-34.
- Uden P (2018). Plant organic acids in fresh and ensiled forage plants. *Grass Forage Science* 73: 583-587.
- Umucalılar HD, Gülşen N, Coşkun B, Hayirli A, Dural H (2007). Nutrient composition of mistletoe (*Viscum album*) and its nutritive value for ruminant animals. *Agroforestry Systems* 71: 77-87.
- Van Soest PJ (1963). The use of detergents in the analysis of fibre feeds. II. A rapid method for the determination of fibre and lignin. *Journal of the Association of Official Analytical Chemists* 46: 829-835.

Van Soest PJ (1967). Development of a comprehensive system of feed analysis and its application to forage. *Journal Animal Science* 26: 119-120.

Yogeshpriya S, Selvara P (2018) Mastery of potassium status and their consequences of hypokalemia in dairy cattle. *Shanlax International Journal of Veterinary Science* 5 (3): 1-5.

Yozgatlı O, Başaran U, Gülümser E, Mut H, Çopur Doğrusöz M (2019). Morphological traits, yield and silage qualities of some corn varieties under Yozgat Ecological condition. *Journal of Agriculture and Nature* 2 (2): 170-177.