Iran Agricultural Research (2023) 42(1) 75-82



Shiraz University

Investigating the physicochemical and fermentation characteristics of the lime pulp silage treated with molasses

Mehran Jalili, Hamid Mohammadzadeh*, Ali Hossein-Khani, Akbar Taghizadeh

Department of Animal Science, Faculty of Agriculture, University of Tabriz, Tabriz I.R. Iran

* Corresponding Author: hamidmhz@tabrizu.ac.ir DOI: 10.22099/IAR.2024.47042.1532

ARTICLE INFO

Article history:

Received 03 April 2023 Accepted 13 January 2023 Available online 13 January 2024

Keywords:

Dry matter digestibility Lime pulp Pectin Ruminants Water holding capacity ABSTRACT - This study aimed to determine the effects of adding molasses on the physicochemical and silage fermentation characteristics, and in vitro ruminal fermentation parameters of fresh and ensiled lime pulps. This research was conducted in the form of a completely randomized design with four treatments including fresh lime pulp without additives (FLP), fresh lime pulp mixed with molasses; (FLPM), fresh lime pulp silage (LPS), and fresh lime pulp silage mixed with molasses (LPMS). Three replications were performed in each treatment Digestibility and gas production were determined using rumen liquid taken from two male Ghezel sheep. The results revealed that the application of molasses had no significant impact on the amount of dry matter. However, a marked distinction (P < 0.05) was observed between the treatments regarding neutral detergent fiber (NDF), acid detergent fiber (ADF), ash, pH, and water holding capacity (WHC). The potential of gas production for FLPM was significantly (P<0.05) higher than that of other treatments. The addition of molasses to fresh lime pulp enhanced (P<0.05) the digestibility of dry matter in the rumen and total digestive tract. However, the presence of molasses in lime pulp silage reduced (P<0.05) the digestibility of dry matter in the rumen and total digestive tract. The results of this study suggested that the use of molasses would enhance fermentation and digestion by altering the composition of pectin in fresh lime pulp. While the lime pulp can be ensiled without molasses, the addition of molasses boosts the nutritive value and digestibility of silage.

INTRODUCTION

The lime tree or Mexican lime (*Citrus aurantifolia*) is a tree that belongs to the *Rutaceae* (citrus) family and the *Rutales* order (Peterson et al., 2006). The fruit on this tree is small and has a thin green skin that turns yellow when fully ripe. Mexican lime is very aromatic, juicy, and acidic (Walheim, 1996). In 2020, Iran ranked 10th in the world with the production of more than 488 thousand tons of lemon and lime (FAO STAT, 2022).

Every year, a significant percentage of limes are processed and separated into the form of fruit juice, essential oil, pulp, and other products (Liu et al., 2022). The residue of lime fruit after juicing is called lime pulp, which mainly includes the peel, core, and pulp residue from juicing. It includes about 54% of the total weight of the fruit, which usually varies depending on the type of fruit, processing methods, and environmental factors (Mahato et al., 2019). Lime pulp is an economical source of sugars and fibers, minerals, and vitamins, phenolic compounds, limonoids and essential oils. These bioactive phytochemicals and other nutrients obtained from lime pulp are nutritious, anti-parasitic, anti-flatulent, antimicrobial, antioxidant, and antiinflammatory. Lime pulp can effectively enhance animal feed quality, improve health, and increase livestock production (Tayengwa et al., 2022).

Considering the seasonal production and low shelf life of lime pulp, the need to use processes that enable the provision of this product throughout the year seems necessary. Storing plant products in the form of silage is a common way to provide food resources for ruminants during a period of the year when fresh fodder is not available (Zinjarde and Gampawar, 2014). In this method, as a result of the activity of lactic acid-producing bacteria and under anaerobic conditions, soluble carbohydrates in fodder water are converted into lactic acid and cause a reduction in pH, thus protecting fodder from microbial spoilage (Filya, 2003).

The main purpose of this study was to analyze the fermentation quality of lime pulp silage. Another aspect that was taken into consideration was the amount of fermentable sugars in the lime pulp. Given its high content of easily fermentable sugars, molasses has become a commonly used ingredient to speed up fermentation and enhance the feeding value (Palmonari et al. 2020; Ke et al. 2023), so the secondary objective of this research was to determine the impact of adding molasses, as a source of supplemental sugars, on the fermentation of mixed silage.

MATERIALS and METHODS

Silage preparation and treatment

The lime pulp was obtained from lemon processing units and was transferred to the Khalat-Pushan laboratory unit of Tabriz University. Treatments included 1- Fresh lime pulp (FLP), 2- Fresh lime pulp mixed with molasses (FLPM, 95:5 ratio on a fresh weight basis), 3- Ensiled lime pulp without additives (LPS), and 4- Ensiled lime pulp mixed with molasses (LPMS, 95:5 ratio on a fresh weight basis).



All treatments were prepared in three replications. For this purpose, laboratory PVC silos with a diameter of 10 cm and a capacity of about six kg of fresh silage were used. These laboratory silos were equipped with a drain valve to drain the leachate. The silos were gradually filled with silage materials and at the same time, pounding and compacting were done. In the end, the lids of the silos were tightly closed by metal fasteners, to ensure that air does not penetrate them.

Chemical and physical analyses

After two months of keeping the silos in the dark and at a temperature of 25 °C, all silos were opened, analyzed and sampled. The levels of pectin, dry matter, ash, neutral detergent fiber (NDF), acid detergent fiber (ADF) in lime pulp were measured prior to and after ensiling. For silages, sampling was performed from a depth of 10 cm of silos. After decanting the silage extract, two samples were taken from each replication. The first sample was used for determining pH, volatile fatty acids, water soluble carbohydrates (WSC) and ammonia-N. Another sample was used to determine chemical composition (dry matter, raw ash, pectin, NDF and ADF).

The dry matter content values of lime pulp and lime pulp silages were determined by incubating them in an oven (60 °C) for 72 h. The NDF and ADF content of the samples were determined using the method of Van Soest (Van Soest et al., 1991). Hemicellulose content was calculated using Equation 1 (Eq 1) as follows: Hemicellulose = NDF – ADF.

Crude ash was determined using incineration of the collected samples in an electric furnace (A.O.A.C, 2005). Water-soluble carbohydrates and ammonia-N were measured by spectrophotometry and volatile fatty acids and lactic acid were measured using high-performance lipid chromatography (HPLC), (Wang et al., 2021). Ammonia concentration in silage extracts was measured for distillation in a Kjeldahl auto analyzer as described by Filya (2003).

To measure pectin, the lime peel powder (10 g) was mixed with hydrochloric acid and citric acid at 200 and 400 g of 0.05 M solution to obtain peel-to-extractant ratios of 1:20 and 1:40, respectively. The mixture was heated at 95 °C with continuous shaking for 1 h in a shaker water bath (Memmert WNB 7–45, Schwabach, Germany). The pectin extract solution was filtrated using a 420 µm screen. The two pectin extract solutions were pooled and centrifuged at 10,000×g (25 °C) for 5 min and then the clear pectin solution was used for subsequent purification.

A one volumetric part of clear pectin solution was precipitated into two volumetric parts of absolute ethanol, stirred for 10 min, and kept for 2 h without stirring at room temperature (25 °C). After filtration, the pectin was washed 3 times with absolute ethanol to remove other components and then finally washed with acetone. The lime peel pectin was left at 50 °C for 1 h using a hot air oven (redLINE RF 115, Deutschland, Germany) to dry off the odorless acetone and then was ground using a Waring blender (HGB2WT, Torrington, CT, USA) and sieved through a 125 µm screen

(Rodsamran and Sothornvit, 2019). The yield of pectin was calculated using the Eq. 2:

Pectin (%) =
$$\frac{Weight \ of \ pectin}{Weight \ of \ Lime \ pulp} \times 100$$
 Eq. 2

Water Holding Capacity (WHC)

To measure the WHC, 2.5 grams of the sample was soaked in 250 ml of distilled water for 24 h. The wet sample was placed on filter paper for 10 minutes, then the sample was weighed (Giger-Reverdin, 2000).

Gas Production Test

The method of Fedorak and Harodi (1983) was employed to gauge the digestibility of organic matter and metabolizable energy through the use of gas production. The amount of gas produced was recorded in hours 2, 4, 6, 8, 12, 24, 36, 48, 72 and 96 of incubation time. The Eq. 3 was used to determine the gas production parameters as described by Ørskov and MacDonald (1979).

Y=b (1-e^{-ct}) Eq. 3 where "Y" (mLg⁻¹ of DM) is gas produced at a time of "t", "b" (mL) is gas production from the fermentable insoluble fraction, "e" otherwise known as Euler's number, is a fundamental mathematical constant that is roughly equal to 2.71828, "c" (mLh⁻¹) is gas production rate for "b" fraction, and "t" (h) is the incubation time.

Digestibility of nutrients

To measure the digestibility of DM, ruminal fluid was collected from two sheep with fistula. The digestibility of the samples was determined using the method of Holden and the Daisy digestion simulator (Holden, 1999).

Statistical analyses

The obtained data were analyzed in a completely random design with four treatments and three replications for each treatment using the SAS statistical program (SAS, 2004). The data were analyzed according to the model $Y_{ij} = \mu + T_i + e_{ij}$. In this model, " Y_{ij} " represents the general observation, " μ " is the overall mean, " T_i " indicates the treatment effect, and " e_{ij} " accounts for the experimental error. Tukey test (at a 5 % probability level) was used to compare the means.

RESULTS and DISCUSSION

The values of ash, NDF, and ADF were significantly (P<0.05) higher in silages containing molasses (LPMS) compared to silages without molasses (LMS) treatment (Table 1). But, the values of HC was significantly (P<0.05) lower in silages containing molasses (LPMS) compared to silages without molasses (LMS) treatment. However, adding molasses did not have a significant effect (P < 0.05) on the dry matter percentage of fresh lime pulp or fresh lime pulp silage (Table 1). Similarly, Kordi et al (2010) used different levels of molasses (0, 6, 12, and 18 g kg⁻¹ fresh citrus pulp) to prepare citrus pulp silage and observed that the percentage of dry matter was not significantly different between treatments. As molasses is one of the most widely used food additives in livestock nutrition, it has been shown that the application of molasses to silage promotes the

access of lactobacilli to soluble carbohydrates, consequently increasing the amount of lactic acid, and reduces the pH of silage (Ke et al., 2023). The results of this research also revealed that molasses can enhance the ash content of silage (Table 1, compare the ash percentage of DM between LPS and LPMS treatments) because of the high mineral content in molasses as discussed by Paviz et al. (2011) and Palmonari et al. (2020).

The values of pH were significantly (P < 0.05) higher in treatments containing molasses compared to treatments without molasses (Table 1). It was shown that the low pH of the fresh lime pulp during ensiling resulted in achieving a suitable silage pH at the end of the period; So, after 60 days of ensiling, the pH of LPS was reported as 3.97 (Table 1). The addition of molasses elevated (P<0.05) the pH value of the fresh lime pulp (4.10 to 4.27) and fresh lime pulp silage (3.97) to 4.21, Table 1). One of the most obvious characteristics of fresh lime pulp in this experiment that sometimes caused a change in the expected results was its initial low pH. After adding five % molasses, fresh lime pulp became alkaline by about 0.2 units. In lime pulp silages, upon the addition of molasses, although the lactic acid content of the silage increased when molasses was used at ensiling, the results revealed an increase in pH value. This may be due to an increase in the percentage of raw ash after adding molasses to the fresh and ensiled lime pulp (Table 1) and also be because of the high macro-minerals (potassium, calcium, etc) and micro-minerals (copper, magnesium, etc) contents in molasses as it has been reported by Palmonari et al. (2020).

It seems that among these minerals, the highest amount of micro-minerals was related to potassium. It has been reported that the potassium content in sugar beet molasses and sugarcane molasses were 1/82 and 2/44 % of their dry matter, respectively (Palmonari et al., 2020). Potassium is an alkaline element that can control pH. Also, potassium specifically helps remove protons or positively charged atoms, which results in a decline in acidity (Aras et al., 2008). Independent studies on the effect of potassium on the pH of animal feed have not been reported, but some studies have suggested that replacing starch with molasses or molasses-based liquid feeds can have positive effects on rumen pH (Oelker et al., 2009; Brito et al., 2017). Further, citrus fruits contain considerable level of natural citrate. The highest level of citric acid has been reported to be in lemon juice (Penniston et al., 2008). Metabolization of citrate to citric acid results in bicarbonate production, which in turn leads to alkalinization of the environment. Minerals in molasses (K, Na, and so on) seem to facilitate this reaction, since citric acid does not become bicarbonate until it is complexed with cations (Gabutti et al., 2009; Aman et al., 2010; Krieger et al., 2015).

Pectin constituted about 43.98% of the dry matter of fresh lime pulp (Table 1). The addition of molasses and ensiling did not cause significant differences in pectin values (Table 1). This finding shows that lactic acid bacteria cannot ferment this carbohydrate. Physicochemical characteristics of lime pulp, whether

fresh or ensiled, are related to its pectin. Pectin-rich cell walls usually have very high hydration properties, which can be attributed to the hydrophilicity and high charge of pectic substances (Thibault and Ralet, 2003). Pectin in lime pulp absorbs water due to its galacturonic acid structure. Pectins are all hydrophilic colloids, so they have a high hydration capacity due to the binding of water molecules to the hydroxyl group of the galacturonic polymethyl chain. In addition, there is a negative charge in the pectin molecule, so they can repel each other and increase the viscosity of the solution. This is because under negative charge conditions and due to high hydration, pectin fibers approach each other, and they interact with each other to create a solid threedimensional network containing a liquid phase inside (Visser and Verragen, 1996). The chemical composition alone cannot adequately reflect the complexity and heterogeneity of lime pulp tissue. According to Table 1, the fresh lime pulp had a high WHC. Adding molasses to a fresh lime pulp lowered its WHC significantly. It seems that the absorption of molasses by lime pulp while completing part of this capacity, by penetrating pectin fibers, reduces the negative charge and hydration due to the interaction with divalent cations, especially Ca²⁺ with high ionic strength, which results in the stabilization of pectin molecule (Castillo-Israel et al., 2015). Also, the abundance of monovalent cations in molasses can influence cell wall swelling due to the known interaction between pectin and monovalent cations, particularly potassium, at a pH below 5 (Yoo et al., 2003). This is because the ionic interactions in the pectic polysaccharide matrix can affect the swelling of the cell wall (Thibault and Ralet, 2003).

Adding molasses to fresh lime pulp lowered its WHC significantly (*P*<0.05). However, no significant difference was observed in WHC between LPS and LPMS indicating that adding molasses to the silage did not affect the WHC of the silage (Table 1). In the experiments of this study, the necessary conditions for drainage and leachate removal were considered for the silos during the 60-days storage period. However, no leachate was extracted from the silos during the entire ensiling period. This finding confirmed the high WHC of lime pulp, which would prevent leachate production even in low dry matter lime pulp (21.69-23.67 %, Table 1).

Regarding of the fermentation the data characteristics (Table 2), the WSC content in LPMS treatment was higher (P<0.05) than that in LPS treatment, likely it is due to the higher total sugar content (62.1% of dry matter on average) found in molasses (Palmonari et al., 2020). The amount of lactic acid in silage containing molasses (LPMS) was also higher (P < 0.05) than that in LPS treatment (Table 2), Also, a higher (P < 0.05) concentration of acetic acid and propionic acid was observed in LPS treatment compared with LPMS treatment. However, in silage containing molasses (LPMS), a significant increase (P<0.05).in the amounts of valeric acid, and isovaleric acid was observed compared with LPS treatment (Table 2).

Adding molasses elevated the final concentration of butyric acid in the lime pulp silages (Table 2). Studies have demonstrated that saccharolytic fermentation is linked to increased production of ketogenic fatty acids (Chen et al., 2010). According to reports, sucrose makes up 48.8% of the dry matter in sugarcane molasses and 60.9% in dry beet molasses (Palmonari et al., 2020). This leads to an increase in butyric acid production in silage due to the high sucrose content in molasses.

There was a significant (P<0.05) difference in gas production values (mLg⁻¹ of DM) among treatments (Table 3). Slowly degraded fraction (b) amount (mL) and rates (mLh⁻¹) of gas production (c) also differed (P<0.05) among treatments. FLPM treatment had the highest "b" and "c" values. Upon adding molasses to fresh lime pulp, significantly (P<0.05) increased the amount of gas production. Similarly, Behasharti et al (2017) observed that adding citrus pulp and application of molasses during alfalfa silage preparation enhanced gas production in a laboratory experiment.

Rumen rapidly and extensively breaks down pectin (Barrios-Urdaneta et al., 2003). *In vitro* gas production is heavily reliant on the amount of fermentable carbohydrates (Behasharti et al., 2017). It has been shown that increasing rumen fermentation is facilitated by pectin in lime pulp and carbohydrates in molasses resulting in increasing gas production (Lashkari and Taghizadeh, 2012; Palmonari et al., 2020).

During the 60-day ensiling period, the fermentation of WSC by Lactobacilli in the LPMS treatment contributed to a decrease in rumen fermentation and gas production (P<0.05), as the number of fermentable carbohydrates plays a crucial role in *in vitro* gas production (Behasharti et al., 2017). The FLPM exhibited the highest gas production rate due to the well-known fact that lime pulp pectin and molasses carbohydrates enhance rumen fermentation. (Lashkari and Taghizadeh, 2012; Palmonari et al., 2020).

Table 1. The data of the chemical and physical properties of this study's treatments including fresh lime pulp without additives (FLP), fresh lime pulp mixed with molasses (FLPM), fresh lime pulp silage (LPS), and fresh lime pulp silage mixed with molasses (LPMS)

Items*	Treatment**				- SEM	<i>P</i> -VALUE
	FLP	FLPM	LPS	LPMS	SEIVI	P-VALUE
pН	4.10 ab	4.27 a	3.97 b	4.21 a	0.061	0.0055
DM (%)	23.67	23.26	21.13	21.69	0.972	0.0771
Ash (% of DM)	6.55^{b}	7.42a	6.63 b	7.93 a	0.032	0.0001
NDF (% of DM)	25.13 ^a	22.73°	23.82 b	24.73a	0.533	0.0001
ADF (% of DM)	19.93a	$18.07^{\rm b}$	18.36 b	19.69 a	0.389	0.0001
HC (% of DM)	5.20^{b}	4.73°	5.46 a	5.04 ^b	0.231	0.0001
Pectin (% of DM)	43.98	42.75	41.58	41.96	0.677	0.1873
WHC (%)	70.30^{a}	45.56 ^b	34.33°	32.59°	3.384	0.0013

^{*}DM, dry matter; NDF, neutral detergent fiber; ADF, acid detergent fiber; HC, hemicellulos; WHC, water holding capacity.

Table 2. The data of the fermentation characteristics of fresh lime pulp silage (LPS), and fresh lime pulp silage mixed with molasses (LPMS)

Item*	Treatment**			P-VALUE	
item.	LPS	LPMS	— SEM	r-value	
WSC (Mmol /100 mol)	2.56 ^b	3.43a	0.142	0.0002	
LA (Mmol/100 mol)	43.31 ^b	54.98a	0.956	0.0066	
AA (Mmol /100 mol)	47.84 ^a	37.14^{b}	1.891	0.0298	
PA (Mmol /100 mol)	4.88 a	2.06 ^b	0.235	0.0024	
BA (Mmol /100 mol)	1.25 ^b	3.42a	0.193	0.0078	
VA (Mmol /100 mol)	0.86^{b}	1.12ª	0.005	0.0004	
IVA (Mmol/100 mol)	0.85^{b}	1.06a	0.025	0.0145	
AA: LA	0.91^{b}	1.48 ^a	0.071	0.0147	
LA: VFA	0.75^{b}	1.22a	0.049	0.0109	

^{*}WSC, water soluble carbohydrates; LA, lactic acid; AA, acetic acid; PA, propionic acid; BA, butyric acid; VA, valeric acid; IVA, isovaleric acid; VFA, volatile fatty acids

^{**}Values sharing the same letter are not significantly (P<0.05) different. SEM, the standard error of the mean. P-VALUE, the probability value.

^{**} Values sharing the same letter are not significantly (P<0.05) different. SEM, the standard error of the mean; P-VALUE, the probability value

Table 3- The data of total gas production (mLg⁻¹ of DM), estimated gas production from fermentable insoluble fraction (mL) and estimated gas production rate (mLh⁻¹) of this study's treatments including fresh lime pulp without additives (FLP), fresh lime pulp mixed with molasses (FLPM), fresh lime pulp silage (LPS), and fresh lime pulp silage mixed with molasses (LPMS)

Time of incubation (h)	Treatments**				CEM	DALLIE
Time of incubation (n)	FLP	FLPM	LPS	LPMS	- SEM	P-VALUE
2	64.53a	68.93ª	63.07ª	51.73 ^b	4.381	0.0001
4	110.93a	116.00a	111.13a	95.69 ^b	4.504	0.0001
6	143.73ab	152.53a	146.36ab	130.42°	4.820	0.0001
8	168.00ab	178.40 ^b	173.96ab	157.35°	6.009	0.0001
12	196.07 ^{ab}	199.40ª	205.44a	187.40 ^b	7.007	0.0001
24	224.27 ^{ab}	235.93a	235.53a	214.73 ^b	7.327	0.0001
36	243.20a	257.86a	253.62a	232.44 °	7.564	0.0001
48	249.87^{ab}	263.33a	258.64a	239.11 ^b	8.018	0.0001
72	251.20ab	265.47a	261.39 ^a	241.96 ^b	8.203	0.0001
96	253.33 ^b	269.87a	265.40 ^a	246.84°	8.262	0.0001
DM degradation characteristics*						
b (mL)	248.00^{b}	262.76a	258.40a	238.74 ^b	8.052	0.0001
c (mLh ⁻¹)	0.127^{a}	0.119^{b}	0.130a	0.127^{a}	0.007	0.0460

^{*}b, gas production from fermentable insoluble fraction; c, gas production rate for "b" fraction.

Table 4 presents the results of examining the digestibility of ensiled lime pulp before ensiling and after 60 days of ensiling in laboratory silos. Holden's method (Holden, 1999) was employed to assess the digestibility of fresh lime pulp and lime pulp silages after incubation. The results revealed that the addition of molasses to fresh lime pulp (FLPM) enhanced

(P<0.05) the digestibility of dry matter in the rumen and total digestive tract. However, the application of molasses in lime pulp silage (LPMS) reduced (P<0.05) the digestibility of dry matter in the rumen and total digestive tract.

Table 4. The data of total tract digestibility of nutrients (%) of this study's treatments including fresh lime pulp without additives (FLP), fresh lime pulp mixed with molasses (FLPM), fresh lime pulp silage (LPS), and fresh lime pulp silage mixed with molasses (LPMS)

Itama*	Treatments				CEM	Р-
Items*	FLP	FLPM	LPS	LPMS	- SEM	VALUE
Total deg (%)	31.15 ^b	34.30 ^a	34.21 ^a	31.16 ^b	1.291	0.0092

^{*}Total deg; Digestibility of the entire digestive system of the feed obtained from Holden's method (Holden, 1999).

CONCLUSIONS

The physicochemical attributes of lime pulp dramatically changed when molasses was added to it, resulting in an increase in digestibility, water-soluble carbohydrates, pH, and ash, and a decrease in water-holding capacity. Ensiling of lime pulp mixed with molasses caused a rise in lactic acid and ketogenic fatty acids. Although lime pulp in its fresh form is more nutritious and digestible, ensiling is a good way to save its main nutrient, pectin, for fermentation in the rumen.

ACKNOWLEDGMENTS

The authors wish to acknowledge the financial support given to this project by the University of Tabriz (Tabriz, Iran).

STATEMENTS & DECLARATIONS

Funding

This work was supported by the University of Tabriz (Tabriz, Iran).

Competing interests

The authors have no relevant financial or non-financial interests to disclose.

Author contributions

All authors contributed to the study's conception and design. Material preparation, data collection and analysis were performed by Mehran Jalili and Hamid Mohammadzadeh. The first draft of the manuscript was

^{**}Values sharing the same letter are not significantly (P<0.05) different. SEM, the standard error of the mean; P-VALUE. The probability value

^{**}Values sharing the same letter are not significantly (P<0.05) different. SEM, the standard error of the mean; P-VALUE, the probability value

written by Mehran Jalili and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Data availability

The datasets generated during the current study are available in the Central Library of Tabriz University-Iran repository.

Ethics approval

This is an in vitro research.

REFERENCE

- Aman,J., Nurmohamedhttps://pubmed.ncbi.nlm.nih.gov/?term=Kalfazade+N&cauthor_id=18946667, S. A, Vervloet, M. G., & Groeneveld, A. B. (2010). Metabolic effects of citrate- vs bicarbonate-based substitution fluid in continuous venovenous hemofiltration: a prospective sequential cohort study. *Journal of Clinical Care*, 25(1), 120-127. https://doi:10.1016/j.jcrc.2009.02.013
- Aras, B., Kalfazade, N., Tuğcu, V., Eray, K., Ozbay, B., Polat, H., & Taşçi, A. I. (2008). Can lemon juice be an alternative to potassium citrate in the treatment of urinary calcium stones in patients with hypocitraturia? A prospective randomized study. *Urological Research*, 36(6), 313-317. https://doi.org/10.1007/s00240-008-0152-6
- A.O.A.C., Association of Official Analytical Chemists. 1990. (2005). Official methodss of analysis. 14th Edition. Washington, DC: A.O.A.C.
- Barrios-Urdaneta, A., Fondevila, M., & Castrillo, C. (2003). Effect of supplementation with different proportions of barley grain or citrus pulp on the digestive utilization of ammonia-treated straw by sheep. *Animal Science*, 76, 309–317.
 - https://doi.org/10.1017/S1357729800053558
- Behasharti, M., Shafipour, N., Abdi, E., & Nemati, Z. (2017). Effects of supplementation alfalfa silage with molasses, orange pulp and *Lactobacillus buchneri* on *in vitro* dry matter digestibility and gas production. *Journal of BioScience and Biotechnology* 6(1), 43-47. Error! Hyperlink reference not valid.
- Brito, A. F., Soder, K. J., Chouinard, P. Y., Reis, S. F., Ross, S., Rubano, M. D & Casler, M. D. (2017). Production performance and milk fatty acid profile in grazing dairy cows offered ground corn or liquid molasses as the sole supplemental nonstructural carbohydrate source. *Journal of Dairy Science*, 100, 8146–8160. https://doi.org/10.3168/jds.2017-12618
- Canvasser, N.E., Rivera, M., Bechis, S., Ingimarsson, J.,
 Knoedler, J., Stern, K., Stoughton, C.L., Wollin, D.,
 Borofsky, M., Bhojani, N., El Tayeb, M., Kamphuis,
 G., Leavitt, D., His, R.S & Scotland, K.M. (2022).
 Over-the-counter alkali agents to raise urine pH and citrate excretion: A prospective crossover study in healthy adults. *Urology*, 168(6), 72-78.
 https://doi.org/10.1016/j.urology.2022.05.049
- Castillo-Israel, K. A. T., Baguio, S. F, Diasanta, M. D. B., Lizardo, R. C. M., Dizon, E. I., & Mejico, M. I. F. (2015). Extraction and characterization of pectin from Saba banana [Musa 'saba' (*Musa acuminata x Musa*

- balbisiana)] peel wastes: A preliminary study. International Food Research Journal, 22(1), 190-195. http://www.ifrj.upm.edu.my/22%20(01)%202015/(29). pdf
- Chen, K., Zhou, J., Liu, L., Liu, J., Du, G., & Chen, J. (2010). Enhancing 2-keto-L-gulonic acid production under hyperosmotic stress by adding sucrose. Chinese Journal of Biotechnology, 26(11), 1507-1513. https://cjb.ijournals.cn/cjben/article/abstract/gc10001507?st=article issue
- FAOSTAT. (2020). Top 20 countries production of lemons and limes. (accessed on 26 November 2022). Retrieved from:
 - http://www.fao.org/faostat/en/#rankings/countries_by_c ommodity
- Fedorak, P. M., & Hrudey, S. E. (1983). A simple apparatus for measuring gas production by methanogenic cultures in serum bottles. *Environmental Technology*, 4, 425-432.
 - https://doi.org/10.1080/09593338309384228
- Filya, I. (2003). The effect of *Lactobacillus buchneri* and *Lactobacillus plantarum* on the fermentation, aerobic stability, and ruminal degradability of low dry matter corn and sorghum silages. *Journal of Dairy Science*, 86, 3575-3581.
- https://doi.org/10.3168/jds.S0022-0302(03)73963-0 Gabutti,
 - L., Lucchinihttps://pubmed.ncbi.nlm.nih.gov/?term=Kalfazade+N&cauthor_id=18946667, B., Marone, G., Alberio, L., & Burnier, M. (2009). Citrate- vs. acetate-based dialysate in bicarbonate hemodialysis: consequences on hemodynamics, coagulation, acid-base status, and electrolytes. *BMC Nephrol*ogy 10, Article number 7.
 - https://doi: 10. 1186/1471-2369-10-7
- Giger-Reverdin, S. (2000). Characterisation of feedstuffs for ruminants using some physical parameters. *Animal Feed Science and Technology*, 86, 53-59. https://doi.org/10.1016/S0377-8401(00)00159-0
- Holden, L. (1999). Comparison of methods of in vitro dry matter digestibility for ten feeds. Journal of Dairy Science, 82, 1791-1794.
 - https://doi.org/10.3168/jds.S0022-0302(99)75409-3 https://doi.org/10.1016/j.juro.2006.11.058
- Ke, W., Wang, Y., Rinne, M., Franco, M. O., Li, F., Lin, Y., Zhang, Q., Cai, Y., & Zhang, G. (2023). Effects of lactic acid bacteria and molasses on the fermentation quality, in vitro dry matter digestibility, and microbial community of Korshinsk peashrub (*Caragana korshinskii* Kom.) silages harvested at two growth stages. *Grass and Forage* Science 2023, 1-13.. https://doi.org/10.1111/gfs.12619
- Kordi, M., Naserian, A. A, Valizadeh, R., & Tahmasbi, A. M. (2010). Effects of different levels of molasses on fermentational properties of citrus pulp silage. Proceeding of 14th Animal Science Congress of AAAP. pp. 198. Taiwan.
- Mahato, N., Sinha, M., Sharma, K., Koteswararao, R., & Cho, M. H. (2019). Modern extraction and purification techniques for obtaining high purity food-grade bioactive compounds and value-added co-products from citrus wastes. *Foods*, 8(11), 523. https://doi.org/10.3390/foods8110523

- Oelker, E. R., Reveneau, C., & Firkins, J. L. (2009). Interaction of molasses and monensin in alfalfa hay- or corn silage-based diets on rumen fermentation, total tract digestibility, and milk production by Holstein cows. *Journal of Dairy Science*, 92, 270–285. https://doi.org/10.3168/jds.2008-1432
- Ørskov, E. R., & McDonald, I. (1979). The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. *The Journal of Agricultural Science*, 92, 499-503. https://doi.org/10.1017/S0021859600063048
- Palmonari, A., Cavallini, D., Sniffen, C. J., Fernandes, L., Holder, L., Fagioli, P. L, Fusaro, I., Biagi, G., Formigoni, A., & Mammi, L. (2020). Characterization of molasses chemical composition. *Journal of Dairy Science*, 103, 6244–6249.
 - https://doi.org/10.3168/jds.2019-17644
- Paviz, M. M., Ghoorchi, T., & Ghanbari, F. (2011). Effect of molasses and bactrial incoulant on chemical composition and aerobic stability of sorghum silage. *Asian Journal of Animal and Veterinary Advances*, 6(4), 385-390. DOI: 10.3923/ajava.2011.385.390
- Penniston, K.L., Nakada, S.Y., Holmes, R.P., & Assimos, D.G. (2008). Quantitative assessment of citric acid in lemon juice, lime juice, and commercially available fruit juice products. *Journal of Endourolog*. 22(3), 567–570. https://doi.org/10.1089/end.2007.0304
- Peterson, J. J., Beecher, G. R., Bhagwat, S. A., Dwyer, J. T, Gebhardt, S. E., Haytowitz, D. B., & Holden, J. M. (2006). Flavanones in grapefruit, lemons and limes: A composition and review of the data from the analytical literature. *Journal of Food Composition and Analysis*, 19, 574-580. https://doi.org/10.1016/j.jfca.2005.12.009https://doi.org/10.1016/j.jfca.2005.12.009
- Lashkari, S., & Taghizadeh, A. (2012). Estimating of chemical composition, degradability, and fermentation parameters of citrus by- products using in situ and gas production techniques. *Journal of Animal Science Research*, 23(1), 15-28. (In Persian).
 - https://animalscience.tabrizu.ac.ir/article_382_85.html? lang=en
- Liu, S., Li, S. h., & Ho, C. T. (2022). Dietary bioactives and essential oils of lemon and lime fruits. *Food Science and Human Wellness*, 11(4), 753-764. https://doi.org/10.1016/j.fshw.2022.03.001
- Rodsamraa, P., & Sothornvit, R. (2019). Microwave heating extraction of pectin from lime peel:

- Characterization and properties compared with the conventional heating method. *Food Chemistry*, 278, 364-372.
- https://doi.org/10.1016/j.foodchem.2018.11.067
- SAS Institute. (2004). SAS®/STAT Software, Release 9.4. Cary, NC. USA: SAS Institute, Inc.
- Thibault, J. F., & Ralet, M. C. (2003). Physico-chemical properties of pectins in the cell walls and after extraction. In Voragen, F., Schols, H., Visser, R. (Eds) *Advances in pectin and pectinase research* (PP 91-105). Dordrecht: Springer. https://doi.org/10.1007/978-94-017-0331-4
- Tayengwa, T., Katiyatiya, C. L. F, Mahachi, L. N., Chikwanha, O. C., & Mapiye, C. (2022). Citrus biowastes: Applications in production and quality enhancement of food from animal sources. In: Ramadan, M. F., Farag, M. A. (eds) *Mediterranean fruits bio-wastes*. (pp. 133-177). Cham: Springer. https://doi.org/10.1007/978-3-030-84436-3
- Yoo, S. H., Fishman, M. L., Savary, B. J., & Hotchkiss, A. T. (2003). Monovalent salt-induced gelation of enzymatically deesterified pectin. *Journal of Agricultural and Food Chemistry*, 51(25), 7410–7417. https://doi.org/10.1021/jf0301520
- Van Soest, P. V., Robertson, J. B., & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science*, 74(10), 3583-3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2
- Visser, J., & Voragen, A. G. J. (Eds.) (1996). Pectins and pectinases. In progress in biotechnology book series. Vol. 14, Pages 3-990.
- Walheim, L. (1996). Citrus: Complete guide to selecting, growing and using more than 100 varieties in California, Arizona, Texas and Florida. USA: Ironwood Press.
- Wang, M., Wang, H., Zheng, H., Uhrin, Dusan., Dewhurs, R. J., & Roehe, R. (2021). Comparison of HPLC and NMR for quantification of the main volatile fatty acids in rumen digesta. *Scientifc Reports*, 11, 24337. https://doi.org/10.1038/s41598-021-03553-9
- Zinjarde, R. M., & Gampawar A. S. (2014). Preservation of orange pomace by ensiling with paddy straw-A laboratory trial. *The Indian Journal of Animal Sciences*, 72(3), 266-268.
 - https://epubs.icar.org.in/index.php/IJAnS/article/view/37505.



بررسی ویژگیهای فیزیکوشیمیایی و تخمیری سیلوی تفاله لیموترش تیمار شده با ملاس

مهران جلیلی، حمید محمدزاده *، علی حسین خانی، اکبر تقیزاده

گروه علوم دامی، دانشکده کشاورزی، دانشگاه تبریز، تبریز، ج.ا. ایران

*نویسنده مسئول

اطلاعات مقاله

تاريخچه مقاله:

تاریخ دریافت: ۱۴۰۲/۰۱/۱۴ تاریخ پذیرش: ۱۴۰۲/۱۰/۲۳ تاریخ دسترسی: ۱۴۰۲/۱۰/۲۳

واژههای کلیدی: پکتین تفاله لیموترش ظرفیت نگهداری آب قابلیت هضم ماده خشک نشخوار کنندگان

چکیده- هدف از مطالعه حاضر تعیین اثرات کاربرد ملاس بر ویژگیهای فیزیکوشیمیایی و تخمير سيلويي و قابليت هضم تفاله ليموترش تازه و سيلو شده بود. اين پژوهش در قالب طرح کاملاً تصادفی با جهار تیمار و سه تکرار در هر تیمار انجام شد. تیمارها شامل تفاله لیموترش تازه بدون مواد افزودني (FLP)، تفاله ليموترش تازه مخلوط با ملاس (FLPM)، سيلو تفاله ليموترش (LPS) و سيلو تفاله ليموترش تازه مخلوط با ملاس (LPMS) بودند. براي بررسي قابلیت هضم و تولید گاز، مایع شکمبه از دو گوسفند نر نژاد قزل گرفته شد. نتایج نشان داد که اثر افزودن ملاس بر میزان ماده خشک معنی دار نبود. با این حال، بین تیمارها از نظر ADF ،NDF و ظرفیت نگهداری آب تفاوت معنی داری (P<-/-۵) وجود داشت. پتانسیل تولید گاز برای FLPM به طور معنی $(P<\cdot/\cdot \Delta)$ بیشتر از سایر تیمارها بود. افزودن ملاس به تفاله لیموترش تازه باعث افزایش ($P < \cdot / \cdot \Delta$) قابلیت هضم ماده خشک در شکمبه و کل دستگاه گوارش شد. در عین حال قابلیت هضم ماده خشک در شکمبه و کل دستگاه گوارش، با افزودن ملاس به سیلاژ لیموترش کاهش داشت $(P<\cdot/\cdot \Delta)$. نتایج این مطالعه نشان داد که ملاس میتواند از طریق تغییر در ساختار پکتین موجود در تفاله لیموترش تازه برای بهبود کیفیت تخمیر و هضم آن مورد استفاده قرار گیرد. تفاله لیموترش را مى توان بدون افزودن ملاس سيلو نمود؛ اما با افزودن ملاس قابليت هضم و ارزش غذايي سيلاژ بهبود مييابد.