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Effects of Raw Potato Waste, Molasses, and Bacterial Inoculation on Chemical Composition, Fermentation Quality, and *in vitro* Gas Production in Corn Silage

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ABSTRACT

This study aimed to assess and compare the impact of adding raw potato waste (**RPW**) and molasses, with and without bacterial inoculations, on the composition, fermentation quality, and *in vitro* gas production in corn silage. The experiments were conducted on 8 treatments: corn silage without additives (**CS**), CS with molasses at 4% (**CSMol**), CS with RPW at 8% (**CSPot8**), and CS with RPW at 10% (**CSPot10**). The remaining four treatments were similar to the above treatments, but with bacterial inoculants added (**CSb**, **CSMolb**, **CSPot8b** and **CSPot10b**). The greatest dry matter, crude protein, and ash as well as the lowest neutral detergent fiber, were observed in the CSMolb. CSPot10 and CSPot8 did not show any significant difference in terms of water-soluble carbohydrates. No significant differences in pH were found among the groups treated with bacterial inoculants and CSMol.The highest concentrations of lactic and propionic acids, as well as the lowest concentrations of butyric acid and Ammonia-N, were all observed in the CSMolb group. Additionally, the lowest acetic acid concentration was detected in CSPot8. Regarding gas production at 24h (**GP24**), the groups that received molasses or RPW (with/without bacterial inoculants) had similar values of GP24, with the only significant difference found in CS and CSb treatments. Overall, CSMolb demonstrated superior performance compared to the other experimental treatments. Additionally, CSPot8b and CSPot10b treatments exhibited favorable fermentation parameters. These findings suggest that RPW can be effectively incorporated as a component of corn silage

Key words: Corn silage, fermentation, gas production, nutritive value, potato waste

INTRODUCTION

Introduction

The rapid population growth in developing countries and the limited supply of animal protein is increasing the demand for livestock products. This need has led to a further increase in food imports from industrialized countries (Tian *et al.* 2016; Henchion *et al.* 2017). Therefore, in order to achieve sustainable agriculture and animal husbandry, it is essential to increase domestic production. To support increased livestock production, the agricultural wastes can be used for feeding livestock (Salemdeeb *et al.* 2017). One potential agricultural byproduct in Iran is raw potato waste (**RPW**), which results from the lack of attention to complete physiological handling of the product at harvesting time, and use of inappropriate methods at harvesting time, grading, transport and packaging. Although production of food from these wastes is possible, the cost of drying and processing make it economically impossible (Nkosi *et al.* 2010). Thus, it seems that these wastes can be used in as feed ingredients for livestock.

Considering the high moisture content of potato waste, it must be preserved in a cost-effective manner so that it can be fed during the year. Ensiling is commonly used to preserve and maintain the quality of feeds with high moisture

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(Chedly and Lee, 2000) that could be used for potato waste. But for a stable fermentation, adequate amounts of fermentable carbohydrates are needed to produce lactic acid to lower the pH. The concentration of water-soluble carbohydrates in potato waste is low (Nkosi and Meeske, 2010), thus ensiling potato waste alone is not possible. Although some additives can be used to improve fermentation, the use of additives has limitations (Nkosi and Meeske, 2010). Alternatively, these wastes can be mixed with other wet feeds such as chopped whole-plant corn that has adequate sugar concentrations for ensiling. Previous research has shown that ensiling materials with or without bacterial inoculant (Blajman *et al.* 2018) and adding molasses (Mordenti *et al.* 2021) can improve the quality of bacterial fermentation and of the silage.

To the best of our knowledge, there is currently no available information regarding the utilization of RPW in combination with corn silage. The objective of this study was to assess and compare the impact of adding RPW and molasses, with or without bacterial inoculation, on the chemical composition, fermentation quality, and *in vitro* gas production in corn silage.

MATERIALS AND METHODS

Whole-plant corn grown in June at a corn field in Semnan, Iran, was used. The field was fertilized with 120 kg of N (as urea) and 40 kg of phosphorous (as triple superphosphate) per ha. Weeds were controlled through row cultivation during the first several weeks after planting. Whole-plant corn was harvested (to a 20-cm stubble height) at the mid-milk stage of kernels by a corn harvester in early autumn. Prior to ensiling, whole corn plants were mechanically cut into approximately 2 to 4 cm particle lengths without undergoing wilting.

Waste from Agria potatoes prepared from a potato farm in Semnan and reduced into small shreds by grating. A total of 400 kg corn whole plant was divided into 8 parts, each assigned randomly to one treatment. The experiment consisted of 8 treatments, four of which had no bacterial inoculants: corn silage without additives (**CS**), CS with molasses at 4% (**CSMol**), CS with RPW at 8% (**CSPot8**), and CS with RPW at 10% (**CSPot10**). The remaining four treatments were similar, but with bacterial inoculants added (**CSb**, **CSMolb**, **CSPot8b** and **CSPot10b**).

Each treatment had five replicates. Each of the four non-inoculated groups received 50 kg forage corn supplemented with molasses or RPW, homogenized, and vacuumed into 5 three-layer 10-kg bags. For the remaining four treatments (**CSb**, **CSMolb**, **CSPot8b** and **CSPot10b**), the above procedure was followed by bacterial inoculation by adding BioStabil Mays (Biomin, Austria). This inoculant contains several bacterial strains including *Enterococcus faecium*, *Lactobacillus brevis*, and *Lactobacilusplantarum*, as well as inulin as carrier. BioStabil Mays was applied at a rate of 400 mL per 100 kg of fresh material (0.4 g of inoculant was dissolved in 400 mL of water) to obtain at least 1 $\times 10^5$ cfu/g of fresh material. The treatments without bacterial inoculate were sprayed with 400 ml of distilled water per 100 kg of fresh forage. The silages were stored at room temperature (28 to 30°C) for 90 d. At opening of each silo (i.e., each replicate), the contents were mixed thoroughly by hand, and then 3 sub-samples of approximately 3 kg each were collected for chemical analyses, determination of fermentation parameters, and *in vitro* gas production.

Samples were oven-dried at 65°C for 48 h and then milled to pass through a 1-mm screen for chemical analyses. Dry matter (**DM**), crude protein (**CP**), ash and ether extract (**EE**) were determined according to AOAC (2005). The water-soluble carbohydrates (**WSC**)was determined using the anthrone method (MAFF, 1982). Analyses of neutral detergent fiber (**NDF**) and acid detergent fiber (**ADF**) contents were conducted as outlined by Van Soest *et al.* (1991). Concentrations of organic acid and Ammonia-N were measured using the method of Babaeinasab *et al.* (2015). The silage was squeezed to obtain silage liquid. Two milliliters of the liquid were pipetted into a micro-centrifuge tube with 0.5 ml of an acid solution (containing 20% ortho-phosphoric acid and 20 mM 2-ethyl butyric acid, as the internal standard), and centrifuged at 15,000 × g for 15 min at a temperature of 4°C. The supernatant was used to determine lactic acid and VFA using gas chromatograph equipped with a flame ionization detector (FID; 250°C), split-injection port (1.0 µL injection), and a capillary column (Agilent J&W HP-FFAP; 10 m, by 0.535 mm, by 1.00 µm. 19095F-121, Agilent Santa Clara, CA). Ammonia-N concentrations were conducted on the extract obtained by squeezing the silage material and was filtered using Whatman 54 filter paper. A 9 ml aliquot was mixed with 1 ml of a 7.2 N H_2SO_4, and stored at -20°C. After thawing, the silage extracts were analyzed for Ammonia-N using a phenol-hypochlorite assay (Broderick and Kang, 1980). The pH was measured by mixing 50 g of the silage liquid with 125 mL of distilled water in a screw capped bottle. The mixed solution was allowed to stand for 1 h at 25°C with occasional stirring. After decanting the silage extract into a beaker, the pH value was recorded (Babaeinasab *et al.* 2015).

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The methods of Menke and Steingass (1988) were employed in determination of *in vitro* gas production from the treatments. Rumen fluid was collected via ruminally cannulated bulls before the morning feeding. The animals were fed a diet based on alfalfa hay and concentrate. Two hundred milligrams of each sample were incubated with 30 mL of buffered rumen fluid in 100-mL glass syringes at 39°C. Three syringes were included as blanks and only contained buffered rumen fluid. The volume of gas produced was recorded after 2, 4, 8, 16, 24, 48, 72, and 96 h of incubation. The values were corrected for the blanks and expressed in mL per 200 mg of DM. Total gas production were fitted to the exponential equation $Y=b(1-e^{-ct})$, where Y is the gas vol at time t, *b* is the asymptotic value of GP (mL/200 mg of DM), and *c* is the first order fractional rate constant of gas production (per hour). The model parameters were estimated by nonlinear regression (**Proc NLIN**) in SAS (Version 9.1). The metabolizable energy (**ME**), organic matter disappearance (**OMD**) content was calculated using the equations developed by Menke and Steingass (1988).

Chemical composition, fermentation quality and *in vitro* characteristics were analyzed using the general linear models (**GLM**) procedure of SAS (Version 9.1) in a completely randomized design (8 treatments × 5 replicates × 3 individual samples). Means were compared for statistical significance using Duncan's multiple range tests. Differences with $P<0.05$ were considered statistically significant.

RESULTS AND DISCUSSION

The chemical compositions of treatments are shown in Table 1. The greatest DM content was found in CSMolb and CSMol, which exhibited a significant difference from other experimental groups, while the lowest DM content was found in CSb and CS ($P<0.05$). The findings indicated that adding bacterial inoculant alone did not influence DM content of the silage, as found for CSPot8, CSPot10, CSPot8b and CSPot10b, with or without bacteria. This finding is in line with Baah *et al*. (2011), who noted that DM content of silage was unaffected when bacteria were added. It should be noted that greater DM content of CSMolb and CSMol was due to greater DM contents of molasses (60.3%). Irrespective of the nature of supplementation, numerous studies have consistently reported higher mean DM content in corn silage when compared to the results obtained in the present study. The DM content observed in our study aligns with the findings of Babaeinasab *et al*. (2015).

Table 1 Chemical composition (% of DM) of treatments

	Treatment[1]									
Item[2]	**CS**	**CSMol**	**CSPot8**	**CSPot10**	**CSb**	**CSMolb**	**CSPot8b**	**CSPot10b**	SEM	P -value
DM	19.33^{e}	21.65^{ab}	20.65^{d}	21.12^{cd}	19.54^{e}	21.76^{a}	20.82^{cd}	21.23^{bc}	0.097	<0.001
CP	8.41^{d}	9.09^{b}	8.39^{d}	8.54^{cd}	8.90^{b}	9.61^{a}	8.84^{bc}	9.13^{b}	0.052	<0.001
NDF	51.94^{a}	48.11^{d}	50.60^{b}	49.35^{c}	49.75^{c}	46.33^{e}	48.11^{d}	48.80^{e}	0.199	<0.001
ADF	29.43^{a}	26.35^{d}	28.54^{b}	27.38^{c}	26.84^{cd}	24.27^{e}	24.51^{e}	22.21^{f}	0.221	<0.001
Ash	8.22^{c}	10.26^{a}	8.24^{c}	8.14^{c}	9.44^{b}	10.53^{a}	9.30^{b}	9.40^{b}	0.089	<0.001
EE	4.05^{a}	3.93^{a}	3.72^{b}	3.63^{b}	4.02^{a}	3.94^{a}	3.73^{b}	3.63^{b}	0.020	<0.001
NFC	27.36^{f}	28.59^{de}	29.03^{cd}	30.32^{b}	27.86^{ef}	29.57^{bc}	29.97^{bc}	31.64^{a}	0.164	<0.001
WSC	1.48^{d}	1.81^{b}	1.95^{a}	2.04^{a}	1.34^{e}	1.55^{cd}	1.63^{c}	1.78^{b}	0.023	<0.001

^{abcdef}The means within the same row with at least one common letter, do not have significant difference (P>0.05).

[1]CS: corn silage, CSMol: CS with molasses at 4%, CSPot8: CS with potato at 8%, CSPot10: CS with potato at 10%, CSb: CS with bacterial inoculants, CSMolb: CS with molasses at 4% and bacterial inoculants, CSPot8b: CS with potato at 8% and bacterial inoculants, CSPot10b: CS with potato at 10% and bacterial inoculants.

[2]DM: dry matter, CP: crude protein, NDF: neutral detergent fiber, ADF: acid detergent fiber, EE: ether extract, NFC: non-fiber carbohydrates, WSC: water-soluble carbohydrates.

This similarity can be attributed to the cultivation of forage corn in Iran as a secondary crop during the summer season, particularly at latitudes where the available sunlight and temperature conditions may not be optimal for achieving the highest levels of maturity. The greatest CP content was found in CSMolb ($P<0.05$) while the lowest was observed in CSPot8, showing no significant difference from CSpot10 and CS. The greater CP content in CSMol can be attributed to a slightly greater CP content of molasses. Protein contents in the groups that received bacteria were slightly greater compared to those without bacteria, probably due to faster fermentation and pH drop which inhibited the activities of protein-decomposing bacteria. The lowest NDF was found in CSMolb, followed by CSPot8b and CSPot10b, due to lower NDF of molasses and RPW compared to forage corn. In general, groups treated with bacteria had lower NDF than groups that were not treated with bacteria, probably due to increased cell wall degradation and bonded NDF hydrolysis caused by greater silage fermentation. In addition, greater sugar contents for groups treated with molasses

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and potato appear to have lowered NDF by increasing fermentation. The greatest and lowest ADF were found in CSPot10b and CS, respectively (P<0.05). The decreasing the ADF trend in the groups receiving molasses, RPW, and bacteria was similar to the decreasing trend for NDF. Li *et al.* (2014) reported that king grass silage containing molasses, glucose, or sucrose resulted in significant decrease in NDF and ADF compared to untreated king grass silage. The greatest ash content was found in CSMolb, and CSMol, while the lowest was in CSPot10, showing no significant difference from CSPot8 and CS. Adding molasses with 11% ash resulted in increased ash content in molasses-treated groups. As ash content of potato is equal to that of forage corn, adding RPW did not directly affect ash content of the raw materials. Regarding EE, CS, CSMol, CSb, and CSMolb were not significantly different from each other. Our analysis revealed that forage corn had EE content of 3.7%, while molasses and RPW contained 0.35% and 0.3% EE, respectively. Therefore, supplementation with molasses lowered silage EE and supplementation with RPW further reduced EE in the experimental groups. Greatest and lowest non-fiber carbohydrates (**NFC**) levels were found in CSPot10b and CS, respectively. Given the formula used to calculate NFC, parallel changes in CP, NDF, and ash during fermentation has led to similar changes in silage NFC. In terms of WSC, no significant difference was observed between CSPot8 and CSPot10, while the lowest WSC content was found in CSb, which was not significantly different from CSMol. In this experiment, forage corn, molasses, and RPW exhibited WSC contents of 16%, 48%, and 60%, respectively. Consequently, the inclusion of RPW and molasses in the silage formulation led to higher WSC levels compared to CS, regardless of the presence or absence of bacteria.

Table 2 The values of pH, total and individual fermentative fatty acids (FFA, g/kg of DM), and Ammonia-N (g/kg of total N)

Item	Treatment[1]								SEM	P-value
	CS	**CSMol**	**CSPot8**	**CSPot10**	**CSb**	**CSMolb**	**CSPot8b**	**CSPot10b**		
pH	3.9^{a}	3.5^{b}	3.9^{a}	3.9^{a}	3.5^{b}	3.4^{b}	3.5^{b}	3.5^{b}	0.01	<0.001
Total FFA	109.1^{d}	119.3^{b}	109.8^{d}	109.6^{d}	116.1^{c}	121.1^{a}	116.7^{c}	120.2^{ab}	0.46	<0.001
Lactic acid	83.7^{c}	89.7^{a}	84^{c}	84.1^{c}	87.2^{b}	90.3^{a}	87.9^{b}	89.6^{a}	0.26	<0.001
Acetic acid	22.8^{d}	26.5^{b}	23.2^{d}	23^{d}	25.8^{bc}	27.5^{a}	25.6^{c}	27.7^{a}	0.20	<0.001
Propionic acid	1.8^{c}	2.6^{ab}	1.8^{c}	1.9^{c}	2.5^{b}	2.8^{a}	2.7^{ab}	2.5^{b}	0.04	<0.001
Butyric acid	0.67^{a}	0.37^{c}	0.63^{a}	0.50^{b}	0.49^{b}	0.33^{c}	0.38^{c}	0.35^{c}	0.013	<0.001
Ammonia-N	22^{a}	15.3^{c}	21.9^{a}	19.6^{b}	19.2^{b}	11.8^{e}	15.5^{c}	14.8^{d}	0.32	<0.001

^{abcd}The means within the same row with at least one common letter, do not have significant difference (P>0.05).
[1]CS: corn silage, CSMol: CS with molasses at 4%, CSPot8: CS with potato at 8%, CSPot10: CS with potato at 10%, CSb: CS with bacterial inoculants, CSMolb: CS with molasses at 4% and bacterial inoculants, CSPot8b: CS with potato at 8% and bacterial inoculants, CSPot10b: CS with potato at 10% and bacterial inoculants.

The fermentation quality of the various treatments is presented in Table 2. With respect to pH levels, there were no significant differences observed among the groups that received both bacteria and CSMol. In addition, pH values for these groups were lower than other experimental groups. This decline in pH can be attributed to the initial rise in lactobacillus populations and the availability of sugar resources. A decrease in pH and an elevation in lactic acid content within silage may be associated with microbial inoculation, as documented by Kung *et al.* (1987) and Aksu *et al.* (2004). In addition, Nkosi *et al.* (2010) reported further reduction in pH and increased concentration of lactic acid as a result of adding *Lactobacillus buchneri* to potato hash TMR silage in comparison to a group that did not receive *Lactobacillus buchneri*. Additionally, Nkosi and Meeske (2010) reported a significant decrease in pH and a rise in lactic acid content in molasses potato hash silage, as opposed to untreated potato hash silage.

The greatest total fermentative fatty acids (**TFFA**)were found for CSMolb, showing no significant difference from CSPot10b while the lowest TFFA was observed for CS, again showing no significant difference from CSPot8 and CSPot10. In general, all bacterial-treated groups had greater TFFA compared to their respective untreated groups. The greatest lactic acid concentration was found for CSMolb, showing no significant difference from CSPot10b and CSMol, while the lowest concentration of lactic acid was found for CS, with no significant difference from CSPot8 and CSPot10. Increased lactic acid concentration in molasses-treated groups (CSMol

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and CSMolb) and CSPot10b was expected due to increased availability of fermentable compounds for the lactobacilli. The ideal lactic acid concentration for corn silage containing 30-40% DM falls within the range of 37-40 grams per kilogram. It is important to note that a decrease in DM content typically leads to an increase in the concentration of lactic acid (Kung *et al.* 2018). The greatest concentration of acetic acid was found for CSPot10b, showing no significant difference from CSMolb. In addition, the lowest acetic acid concentration was found for CSPot8, which was not significantly different from CSPot10 and CS.

All bacteria-treated groups had greater concentrations compared to their respective untreated groups. In a study conducted by Kung *et al.* (2018), the concentration of acetic acid in corn silage was observed to range from 10 to 30 grams per kilogram of DM, which aligns with our own findings. This concentration range can serve as a valuable means to control the growth of fungi, especially when silage is exposed to air. Additionally, it's worth noting that this level of acetic acid can be absorbed through the rumen wall, contributing to the processes of body and milk fat synthesis (Kung *et al.* 2018). All bacteria-treated groups had greater concentrations of propionic acid compared to their respective bacterial-untreated groups. The greatest concentration of propionic acid was found for CSMolb, showing no significant difference from CSMol and CSPot8b. On the other hand, the lowest propionic acid concentration was observed for CS and CSPot8. The lowest concentration of butyric acid was found for CSMolb with no significant differences from CSMol and CSPot8b and CSPot10b.

The greatest butyric acid concentration was found for CS, showing no significant difference from CSPot8. Clostridium activities in silages can produce small amounts of propionic and butyric acids (Broderick and Kang, 1980). Nevertheless, in our experiment, the concentrations of propionic acid and butyric acid exhibited variations falling within the ranges of 1.8-2.8 grams per kilogram of DM and 0.33-0.67 grams per kilogram of DM, respectively. Ammonia-N concentrations in all experimental groups remained below 100 grams per kilogram of DM, indicating acceptable silage quality (McDonald *et al.* 2002). The lowest concentration of Ammonia-N was detected in the CSMolb group, which can be attributed to limited proteolysis, likely influenced by the lower pH observed in this particular group. Conversely, the greatest concentration of Ammonia-N was noted in the CS group. The decreased concentrations of Ammonia-N in the groups that received bacterial inoculation, molasses supplementation, or a combination of both can be attributed to a reduction in proteolysis. This reduction is likely due to the increased concentration of water-soluble carbohydrates (**WSC**) and the faster colonization of bacteria (Hashemzadeh-Cigari *et al.* 2011).

Table 3 *In vitro* ruminal gas production and estimated parameters

Item[2]	Treatment[1]								SEM	P-value
	CS[1]	**CSMol**	**CSPot8**	**CSPot10**	**CSb**	**CSMolb**	**CSPot8b**	**CSPot10b**		
24-h incubation										
GP24	54.16^{b}	54.95^{a}	54.86^{a}	55.19^{a}	54.03^{b}	55.09^{a}	55.24^{a}	55.34^{a}	0.07	<0.001
OMD	721.73^{d}	745.08^{b}	727.97^{c}	730.91^{c}	730.71^{c}	750.39^{a}	740.24^{b}	743.11^{b}	1.01	<0.001
ME	10.23^{e}	10.41^{ab}	10.32^{cd}	10.38^{bc}	10.26^{de}	10.49^{a}	10.42^{ab}	10.47^{a}	0.01	<0.001
96-h incubation										
b	52.63^{d}	78.98^{a}	58.20^{c}	62.14^{c}	61.48^{c}	81.42^{a}	71.88^{b}	77.94^{a}	1.07	<0.001
c	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.001	<0.001

^{abcde} The means within the same row with at least one common letter, do not have significant difference (P>0.05).

[1]CS: corn silage, CSMol: CS with molasses at 4%, CSPot8: CS with potato at 8%, CSPot10: CS with potato at 10%, CSb: CS with bacterial inoculants, CSMolb: CS with molasses at 4% and bacterial inoculants, CSPot8b: CS with potato at 8% and bacterial inoculants, CSPot10b: CS with potato at 10% and bacterial inoculants.

[2]GP24: *in vitro* ruminal gas production at 24 h (mL/200 mg of DM), OMD: organic matter disappearance (g/kg of OM), ME: metabolizable energy (MJ/kg of DM), *b*: the asymptotic value of gas production, *c*: the first order fractional rate constant of gas production.

Regarding gas production at 24 hours(**GP24**), the experimental groups that received either molasses or potato (with or without bacterial inoculation) generated comparable amounts of gas. The notable increase in GP24 resulting from the supplementation with soluble sugars, whether from molasses or potato, is in line with the observations reported by Rezaei *et al.* (2009) and Makkar (2010). The supplementation of bacteria did not have an effect on the volume of gas produced at 24 h, which aligns with the findings reported by Babaeinasab *et al.* (2015) and Contreras-Govea *et al.* (2011). All the groups treated with bacteria exhibited higher OMD compared to their respective groups without bacteria. The highest OMD was observed in the CSMolb group ($P< 0.05$), while the lowest OMD was recorded in the CS group (Table 3; $P<0.05$). The increased levels of OMD in the experimental groups can be linked to the rising trend of total

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volatile fatty acids (McDonald *et al.* 2002). The highest concentration of ME was detected in the CSMolb group, and this result did not significantly differ from CSPot8 and CSPot10. Conversely, the lowest ME concentration was observed in the CS group, and there was no significant difference when compared to CSb. The increased ME concentrations in the experimental groups could be attributed to higher CP contents and greater amounts GP24.In a similar vein, Li *et al.* (2014) reported the lowest ME concentration in the non-supplemented king grass groups, while the molasses-treated groups in their study exhibited the highest ME concentration. The highest *b* fraction was observed in the CSMolb group, and this result did not show a significant difference from CSMol and CSPot10b. Conversely, the smallest b fraction was identified in the CS group. The greater *b* fraction in the molasses-treated group is likely associated with the higher potentially-digestible fraction present in molasses compared to CS or RPW. In accordance with these findings, Li *et al.* (2014) also reported a higher b fraction in king grass silage supplemented with molasses compared to other groups supplemented with sucrose, glucose, or cellulose. No significant difference was observed among the experimental groups in terms of decomposition rate (*c*) ($P > 0.05$). Similarly, in the study by Li *et al.* (2014), the decomposition rate of king grass was not influenced by supplementation (with molasses, sucrose, glucose, or cellulose).

CONCLUSION

The results suggest that CSMolb outperformed the other experimental groups in various aspects, including chemical composition, silage fermentation quality, and gas production. Parameters related to silage fermentation, such as higher lactic acid concentration and lower levels of butyric acid and Ammonia-N, as well as estimated parameters like ME, and OMD, for CSPotb (8% and 10%) were superior to those of un-supplemented corn silage. Consequently, it can be concluded that RPW can be effectively incorporated into corn silage to enhance its overall quality.

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CONFLICT OF INTEREST

There was no conflict of interest.

AUTHOR CONTRIBUTIONS

All authors contributed to the study conception and design. Material preparation and data collection were performed by Fatemeh Kashefi, Ali Mahdavi, and Atta Mahdavi. Data analysis was performed by Ashkan Jebelli Javan, Ata Mahdavi, and Babak Darabighane. The first draft of the manuscript was written by Ali Mahdavi, Ata Mahdavi and Babak Darabighane. All authors read and approved the final manuscript.

DATA AVAILABILITY

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ETHICAL CONSIDERATIONS

Ethical approval is not applicable for this study as it involves exclusively *in vitro* experiments.

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