International Journal of Recycling of Organic Waste in Agriculture (2022) 11: 541-552
Doi:10.30486/IJROWA.2022.1933535.1279

ORIGINAL RESEARCH

Ensiling characteristics of prickly pear (opuntia-ficus indica) rejects with and without molasses for animal feed

L El Hajji [1*], H Azzouzi [1, 2], M Achchoub [1, 2], K Elfazazi [2], S Salmaoui [1]

Received: 26 June 2021 / Accepted: 05 March 2022 / Published online: 01 May 2022

Abstract

Purpose The aim of this work was to study the effect of adding sugar beet molasses on the biochemical properties, microbial flora, fermentation quality, and aerobic stability of prickly pear cactus (*Opuntia ficus-indica*) waste silage.

Method Molasses (0%, 2%, 4%, 6%, 8% and 10%, w/w) was mixed with the cactus fruit scraps, straw and wheat bran.

Results The dry matter content, pH, total and reducing sugars of the pre-ensiling material increased after adding different percentages of the beet molasses ($P < 0.05$). During fermentation, we observed substantial protein and sugar degradation. All silage treatments reached stable pH values (pH 4.3-4.6). Among all the concentrations, the 10% beet molasses treatment underwent the highest lactic acid fermentation. Accordingly, the pH drop was higher in the 10% concentration (1.13 units) compared to lower beet molasses concentration (1.03 units). Also, the 10% concentration has the highest number of lactic acid bacteria. The number of yeast and total aerobic mesophiles decreased continuously during silage. Moreover, during post-fermentation testing, the yeast multiplied little for the 10% concentration of beet molasses.

Conclusion The results show that the addition of molasses has a significant effect on silage characteristics of prickly pear cactus.

Keywords Silage, Fermentation, Cactus rejects, Molasses

Introduction

Livestock farming is one of the key sectors of Moroccan agriculture, generating more than 40% of the annual agricultural turnover (Tazi et al. 2014). Consequently, the demand for livestock feed increases considerably every year, the major constraint is the lack of green fodder for animal feed. Legumes are considered a promising source of protein for livestock nutrition, but their high cost and seasonality make them unavailable to most livestock farmers. In this context, we have focussed on a residue available in significant quantities in Morocco: prickly pear cactus (*Opuntia ficus-indica*) fruit remnants. In Morocco, the cactus plantation has tripled from almost 45.000 ha at the beginning of 1990, to ~ 150.000 ha

✉ L El Hajji elhajjiloubna.1989@gmail.com

1 Ecology and sustainable development Laboratory, Faculty of Science and Technology, Sultan Moulay Slimane University, Beni Mellal, Morocco

2 Laboratory of Food Science and Technology, Regional Center for Agricultural Research in Tadla (CRRAT), Beni Mellal, Morocco

in 2016 (Mabrouk et al. 2016). The cactus is a seasonal product, producing large quantities of fruit over a short time. The fruit maturity period is relatively short, especially when high temperature accompanies the summer season. As a result, between 30 and 50% of the cactus fruits ripens quickly and becomes unfit for human consumption (Bendaou 2013, Ait-Oubahou and Bartali 2015). The cactus fruit is rich in nutrients such as sugars, minerals, and fibers (El Hajji and Salmaoui 2020), therefore it is essential to use these materials in animal feed. The cactus is an important forage in multiple arid and semi-arid regions of the world (Ben Salem et al. 2002). Livestock farmers use it as a support fodder to contrast the frequent periods of drought, which could have disastrous consequences (Le Houérou 1992; Nefzaoui 2000). Its use in livestock feed has many advantages, as it is widespread. Furthermore, it grows rapidly, is an inexpensive crop, is fairly palatable, can withstand long periods of drought (Shoop et al. 1977), has a high biomass yield, and withstands soil salinity (Nobel 2002). In addition, these fruits can be used as fodder, either fresh or preserved as silage (Castra et al. 1977). Ensiling is an anaerobic process for preserving wet crops by lactic fermentation. Under optimal silage conditions, lactic acid bacteria primarily ferment soluble carbohydrates and produce lactic acid, which acidifies the crop and minimizes the activity of aerobic microorganisms, thereby preserving nutrients in the forage (McDonald et al. 1991; Cai et al. 1999). Most of the research on this topic has investigated prickly pear cladodes (Mokoboki et al. 2016). In the current research, we were interested in cactus fruits left in the fields after the harvest. The objective of this study is to explore the possibility of using prickly pear cactus fruit scraps in animal feed using silage as a preservation method and to evaluate the effect of adding increasing levels of molasses on biochemical, microbiological, and fermentation characteristics and aerobic stability of the silage.

Material and methods

Sample collection

Prickly pear cactus samples were collected at the end of the season in the Béni Mellal region (central Morocco) in October 2020, when the fruits were left in the fields after harvest. Samples were immediately chilled (-4°C) after collection.

Silage preparation

Silage was prepared by grinding all cactus fruit scraps. The resulting grist was then added to a straw and wheat bran (whose proportions are 75% of cactus, 12.5% of wheat bran and 12.5% of wheat straws) which acts as a moisture absorbent matrix. The silage was chopped finely to promote compaction of the mixture and to evacuate as much air as possible from the bags before sealing them. To prepare 6 treatments, the mixture was then separated into six portions so that molasses could be added in different percentages. Molasses was then added in the following percentages: 0%, 2%, 4%, 6%, 8% and 10% (w/w). The mixtures were then placed in plastic bags (1 kg/bag) (270*390 mm); Eight bags per treatment. The bags were sealed, lined, and stored for 30 days at room temperature. For pH and microbiological analysis, five bags of each treatment (1 bag/day) were sampled on days 5, 10, 15, 20, and 25. Fresh and silage mixtures had been subjected to analysis of aerobic stability, physicochemical properties, and microbial analysis.

Aerobic stability of silage

After 30 days of silage storage, three bags from each treatment were opened and exposed to the ambient air to study the changes that occurred in the silage during aerobic exposure. The parameters measured were core temperature, pH and yeast counts. These parameters were measured daily for one week.

Chemical analyses

The dry weight was determined by oven drying at 105°C to constant weight, and ash was measured by incineration at 550°C.Crude protein was assessed by the Kjeldahl method described by APHA (1989), fiber was determined by the Van Soest et al. (1991) method, reducing sugars by the Bertrand method (1906), and total sugars by the Dubois et al. (1956) method. The pH was determined using a pH meter after mixing 20 g of the sample in a blender with 50 ml distilled water until a fluid suspension was obtained (Habibi 2004). Elemental analyses (Ca, Fe, Mg, K, Na, and Cu) were by atomic absorption spectroscopy. Fermentation losses were evaluated according to the weight loss expressed in %. All chemical analyses are presented on a DM basis (except DM and pH).

Microbiological analyses

The microbiological characterization was carried out by culturing samples in selective media, as described by Leininger (1976). Plate count agar was used to determine total aerobic mesophilic flora (TAMF) after incubating at 30°C for 72 h. Samples were incubated with potato extract for five days at 25°C to measure yeasts and molds. Lactic acid bacteria were determined after 72 h at 37°C using de Man Rogosa and Sharpe agar. Deoxycholate agar was used to determine total and fecal coliforms, incubated for 24 h at 37°C and 24 h at 44°C, respectively. *Staphylococci* were counted on Baird Parker agar after 48 h at 37°C, *E. coli* on MacConkey agar after 24 h at 37°C, and *Salmonella spp*. on salmonella-shigella agar after 48 h at 37°C. All microbial data were converted to $\log_{10}$ and presented on a fresh matter basis.

Statistical analyses

Statistical analyses were performed using SPSS version 20. The effect of treatment was analyzed using a unidirectional analysis of variance with treatment as the main effect. When measurements were performed on the same sample at different times, the treatment effect was analyzed in a mixed model with treatment, time, and interaction of treatment and time as the main effects. The results are presented for each sample as a mean and standard deviation. For all statistical tests, significance was assigned at $P < 0.05$. All experiments were replicated three times.

Results and discussion

Characterization of raw materials

Cactus rejects is rich in sugar, moisture and minerals; it has medium fiber contents, and low proteins and dry matter concentration. Beet molasses contains mainly sugars and mineral elements. Straw and wheat bran are rich in fiber (Table 1).

Pre-ensiling characteristics of prickly pear waste

The biochemical characteristics of the initial mixtures are presented in Table 2. The initial pH of the first two treatments (0% and 2%) differed significantly from the other treatments. The inclusion of molasses increased the dry matter content ($P < 0.05$); the difference was observed between the 0%, 2% and 10% molasses treatments, due to the high dry matter content of molasses (73.16%). Reducing sugar and total sugar contents increased with molasses addition, likely due to the high sugar content in molasses. These results are consistent with those of Hinds et al. (1985), Lattemae et al. (1996), and Shahsavan (2009). Crude protein decreased with the addition of molasses from 11.67% to 7.00%, 5.83%, 4.08%, 4.67%, and 4.08% DM for the 2%, 4%, 6%, 8%, and 10% treatments, respectively, due to the low protein content of molasses.

The proportion of fiber types (NDF, ADF, ADL, cellulose, and hemicellulose) did not depend on the addition of molasses as molasses does not contain fiber.

Therefore, the differences noted between the treatments depended on the initial composition of initial ingredients (cactus, straw and wheat bran).The mixture without molasses (0%) was rich in calcium (33.52 mg/100 g DM), potassium (382.15 mg/100 g DM), and magnesium (117.97 mg/100g DM). Iron and copper existed in trace amounts (1.06 mg/100g DM and 0.66 mg/100g DM, respectively). These results show that the characteristics of initial mixture are affected by the nutritional value of the initial ingredients (cactus). In this context, Stintzing et al. (2001) and Piga (2004) reported that the cactus was rich in magnesium and calcium, while other minerals were in normal range. The addition of molasses significantly ($P < 0.05$) increased the content of mineral elements such as Ca, Mg, Na, K, Fe, but the Cu content was not affected ($P > 0.05$). In the mixture with 10% molasses, mineral concentrations were 380.58 mg/100g DM for calcium, 137.11 mg/100g DM for magnesium, 11.47 mg/100g DM for sodium, 426.03 mg/100g DM for potassium, and 1.54 mg/100g DM for iron. Thus, in addition to a rich source of carbohydrates, molasses is also a source of minerals.

Table 1 The biochemical composition of raw materials (DM basis %)

Parameters	Cactus scraps	Molasses	Wheat straw	Wheat bran
pH	6.65 ± 0.10	6.00 ± 0.20	-	-
DM	19.85 ± 0.86	73.16 ± 0.15	91.18 ± 1.44	87.66 ± 0.56
Ash	6.5 ± 0.45	12.91 ± 1.08	4.27 ± 0.94	5.75 ± 1.11
Proteins	11.98 ± 0.11	3.12 ± 0.26	2.63 ± 0.88	10.41 ± 1.52
Total sugar	32.34 ± 0.86	74.72 ± 1.02	1.33 ± 1.15	5.82 ± 0.55
NDF	27.23 ± 2.30	0	72.79 ± 1.57	44.69 ± 1.34
ADF	15.67 ± 1.54	0	42.93 ± 0.98	11.65 ± 1.38
ADL	7.11 ± 1.61	0	7.72 ± 1.28	5.40 ± 1.63
Hemicellulose	11.56 ± 3.84	0	29.85 ± 1.77	33.03 ± 2.60
Cellulose	8.55 ± 2.77	0	35.21 ± 1.09	6.25 ± 1.89
Ca (mg/100g)	190.63 ± 8.23	454.59 ± 2.93	48.41 ± 2.25	64.63 ± 0.47
Na (mg/100g)	14.07 ± 2.25	15.17 ± 2.16	1.22 ± 1.22	2.05 ± 0.49
Mg (mg/100g)	25.91 ± 0.86	246.11 ± 0.92	103.16 ± 0.76	94.38 ± 1.16
K (mg/100g)	263.09 ± 1.76	582.51 ± 2.99	128.93 ± 1.81	105.73 ± 2.88
Fe (mg/100g)	0.51 ± 0.04	2.69 ± 0.09	11.32 ± 0.12	7.86 ± 0.36
Cu (mg/100g)	0.51 ± 0.06	2.05 ± 0.04	0.18 ± 0.04	1.07 ± 0.11

DM: Dry matter; FM: Fresh matter; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin

Ensiling of prickly pear waste with molasses

The biochemical characteristics of the mixtures after silage are presented in Table 3.After silage, the DM content increased significantly ($P < 0.05$), likely due to the loss of water in the form of silage effluent. his result is consistent with McDonald et al. (1991), who found that there was slight weight loss at the end of silage. A significant portion of ash was also lost during silage making, which can be explained by its water solubility and loss as effluent. The treatment with 10% molasses showed the highest weight loss, 9.88% of all treatments (which lost between 6.29% and 6.65%).

Table 2 The biochemical composition of the initial mixture (DM basis %) (n = 3)

Parameters	0%	2%	4%	6%	8%	10%	P-value
pH	5.39^{a} ± 0.095	5.39^{a} ± 0.132	5.62^{b} ± 0.115	5.74^{b} ± 0.049	5.74^{b} ± 0.131	5.74^{b} ± 0.130	0.021
DM	31.20^{a} ± 1.868	32.35^{a} ± 1.77	33.92^{ab} ± 2.66	34.46^{ab} ± 2.72	34.61^{ab} ± 0.69	37.72^{b} ± 1.35	0.012
Ash	4.039^{a} ± 0.825	3.021^{a} ± 0.680	4.356^{a} ± 0.505	4.079^{a} ± 0.429	2.670^{a} ± 0.304	3.451^{a} ± 0.865	0.204
Proteins	11.67^{c} ± 1.34	7.00^{b} ± 2.32	5.83^{ab} ± 0.51	4.08^{a} ± 0.51	4.67^{a} ± 0.51	4.08^{a} ± 0.51	0.000
Total sugars	16.97^{a} ± 1.66	20.39^{a} ± 1.01	24.83^{b} ± 10.98	32.32^{c} ± 0.76	37.33^{d} ± 2.28	39.69^{d} ± 0.25	0.000
Reducing sugars	4.78^{a} ± 0.54	10.50^{b} ± 1.02	13.96^{ab} ± 1.07	16.08^{b} ± 0.70	17.69^{b} ± 1.43	22.07^{c} ± 3.34	0.000
NDF	42.09^{b} ± 0.71	50.68^{d} ± 2.54	48.94^{d} ± 1.02	43.78^{b} ± 1.50	37.42^{a} ± 1.13	37.78^{a} ± 0.52	0.000
ADF	20.74^{a} ± 3.04	21.81^{a} ± 1.03	30.13^{b} ± 2.59	20.05^{a} ± 3.38	19.88^{a} ± 3.81	26.20^{ab} ± 1.66	0.032
ADL	15.88^{a} ± 2.81	14.43^{a} ± 2.60	20.90^{a} ± 4.00	13.83^{a} ± 1.22	12.22^{a} ± 3.16	11.11^{a} ± 0.92	0.064
Hemicellulose	21.35^{bc} ± 2.61	28.87^{c} ± 3.57	18.81^{ab} ± 3.61	23.74^{bc} ± 4.84	17.54^{ab} ± 4.53	11.59^{a} ± 1.14	0.016
Cellulose	4.86^{a} ± 2.56	7.39^{a} ± 2.64	9.23^{a} ± 5.18	6.21^{a} ± 3.38	7.67^{a} ± 1.16	15.08^{a} ± 2.08	0.124
Ca (mg/100g)	333.52^{a} ± 2.97	345.19^{b} ± 2.06	351.72^{c} ± 2.28	365.13^{d} ± 1.60	374.74^{e} ± 1.83	380.58^{f} ± 1.14	0.000
Fe (mg/100g)	1.06^{a} ± 0.04	1.17^{ab} ± 0.02	1.23^{bc} ± 0.02	1.30^{c} ± 0.02	1.32^{c} ± 0.01	1.54^{d} ± 0.10	0.000
Mg (mg/100g)	117.97^{a} ± 1.25	121.02^{b} ± 0.23	129.97^{c} ± 0.49	131.69^{d} ± 0.70	135.55^{e} ± 0.54	137.11^{e} ± 0.44	0.000
K (mg/100g)	382.15^{a} ± 5.00	396.77^{b} ± 1.75	403.52^{b} ± 2.00	419.28^{c} ± 3.25	423.41^{c} ± 2.75	426.03^{c} ± 1.75	0.000
Na (mg/100g)	8.29^{a} ± 0.12	9.53^{b} ± 0.06	9.53^{c} ± 0.08	10.83^{d} ± 0.10	10.98^{d} ± 0.11	11.47^{e} ± 0.17	0.000
Cu (mg/100g)	0.66^{a} ± 0.12	0.86^{a} ± 0.05	0.86^{a} ± 0.05	0.88^{a} ± 0.07	0.93^{a} ± 0.08	0.93^{a} ± 0.11	0.088

Values for the same variable with different letters are significantly different. DM: Dry matter; FM: Fresh matter; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin

The pH of all silage treatments was 4.3 to 4.6, meaning that all treatments had good silage quality. McDonald et al. (2002**)** reported that the silage pH between 5 and 7 results in poorly preserved silage. The most significant difference between before and after silage was the sugar content (total sugar and reducing sugar). In all six treatments with and without molasses added, almost 50% of the sugar content was degraded. The higher the proportion of initial sugars, the higher the remaining content. During silage making, sugars are widely used by microorganisms (Jaurena and Pichard 2001). The protein content decreased for all treatments after silage (e.g for the 0% treatment the protein content decreased from 11.67 to 3.5 % DM and from 3.5 to 0.88 % DM for the 10 % treatment), which can be explained by the proteolysis during fermentation. Our results agree with previous studies by Bilal 2009, Moore and Kennedy 1994 and Ni et al. 2017. In these studies authors reported that the addition of molasses to silages decreases the protein content. However,

other researchers (Aksu et al. 2006; Kennedy 1990; Lattemae et al. 1996; McDonald et al. 1991; Mokoboki et al. 2016) reported that the addition of molasses to silages increased crude protein, while Spoelstra et al. 1990 affirmed that the molasses addition did not affect protein content. Acid detergent fiber values are important because they are related to an animal's ability to digest forage. During ensiling, hemicellulose can be hydrolyzed, and types of lactic acid bacteria can ferment pentoses into lactic and acetic acid (McDonald et al. 2002). Although the different treatments significantly affected NDF, ADF, ADL, hemicellulose, and cellulose, their contents generally varied widely. Notably, NDF content decreased after ensiling in the 0%, 2%, 4%, and 6% treatments. In addition, ADF and ADL content also decreased for all treatments. This decrease is probably due to cell wall degradation by plant enzymes or acid hydrolysis (McDonald et al. 1991). Regarding mineral elements, the six treatments generally experienced a substantial loss. Many researchers have reported the successful use of molasses for forage silage (Wuisman et al. 2006; Shellito et al. 2006). Molasses as a sucrose supply also increases the lactic acid bacteria content as well as the lactic acid, and lactic acid is generally the main reason for low pH in high-quality silage. The addition of sucrose to forage legume silages could increase lactic acid production, decrease pH, and improve aerobic stability during storage (Heinritz et al. 2012). Table 4 shows the static mixed ANOVA analysis of the nutritional values, obtained by applying the factor degree of molasses addition, fermentation time, and their interaction. Molasses addition and fermentation time significantly affected dry matter, pH, crude proteins, sugars, NDF, ADF and ADL fibers, and mineral elements (Ca, Fe, Mg, Na, K, Cu). The addition of molasses did not affect the ash content ($P > 0.05$), while the time of silage did not affect the cellulose and hemicellulose contents ($P > 0.05$). The interaction between the two factors did not affect pH, dry matter, ash, ADL, cellulose, or hemicellulose fibers.

Fermentation characteristics

The changes in the fermentation characteristics—pH, lactic acid bacteria, yeast, total aerobic mesophilic flora and total coliforms—during the silage process are shown in Figs 1, 2, 3, 4, 5 respectively. The pH value is commonly used as a criterion for assessing silage quality, and pH values below 4.5 can be considered appropriate (Pettersson 1988). Yang et al. (2004) showed that high humidity > 70% and pH > 4.5 promotes clostridial fermentation. Moreover, pH values decreased during fermentation of all treatments just after the start of fermentation (Fig.1). The pH of the 0%, 2%, 4%, 6%, and 8% treatments was below 4.5 (considered below the pH of microbiological stability in which no microorganisms can grow) during the first five days of ensiling, whereas the 10% treatment did not reach that pH until the 15th day of ensiling. At 30 days of fermentation, therewere three pH values: silage with 0%, 2%, 4% molasses content had a pH of 4.3, silage with 6% and 8% had a pH close to 4.5, and silage with 10% molasses content had a pH close to 4.6. All silages, including the control, appeared to be of good quality, as evidenced by the rapid drop in pH (Fig.1) and low terminal pH. There was a significant increase in lactic bacteria in the first five days of fermentation for all silage treatments (Fig.2).

This result strongly correlates with the drop in pH during this period. From the 6^{th} to the 15^{th} day of fermentation, there was a significant decrease in lactic acid bacteria for all silage treatments, and after the 15^{th} day, their number remained nearly stable. The higher the proportion of molasses, the more intense the lactic acid fermentation and the higher the number of lactic acid bacteria. In other words, this result indicates that lactic acid bacteria consumed more sugar when sugar was available in abundance. A possible explanation for this observation could be that molasses provided sufficient substrate for the growth of lactic acid bacteria in the silage, thus accelerating the production of lactic acid, lowering the pH, and resulting in better fermentation

quality. This phenomenon is confirmed by the results of McDonald et al. (1991).

Table 3 Nutritional assessment after silage (DM basis %) (n = 3)

Parameters	0%	2%	4%	6%	8%	10%	P-value
pH	4.36^{a} ± 0.05	4.36^{a} ± 0.02	4.39^{b} ± 0.01	4.54^{c} ± 0.02	4.54^{c} ± 0.02	4.61^{d} ± 0.015	0.000
DM (%)	31.80^{a} ± 2.17	33.00^{ab} ± 1.06	34.96^{ab} ± 0.41	36.57^{bc} ± 1.81	37.30^{bc} ± 2.12	39.22^{c} ± 0.18	0.011
Ash	1.955^{a} ± 0.027	2.446^{ab} ± 0.364	2.537^{ab} ± 0.384	3.026^{b} ± 0.439	2.229^{ab} ± 0.316	2.243^{ab} ± 0.552	0.244
Proteins	3.50^{d} ± 0.88	2.33^{c} ± 0.51	2.04^{bc} ± 0.51	2.63^{cd} ± 0.88	1.17^{ab} ± 0.51	0.88^{a} ± 0.00	0.002
Total sugars	7.980^{a} ± 0.45	11.18^{ab}± 1.18	13.30^{bc} ± 1.20	16.42^{b} ± 1.40	18.58^{cd} ± 0.46	23.51^{e} ± 1.46	0.000
Reducing sugars	1.86^{a} ± 1.24	2.33^{a} ± 2.18	1.86^{a} ± 1.55	3.26 ^{a} ± 0.62	4.43^{a} ± 2.18	4.43^{a} ± 2.64	0.66
NDF	38.55^{a} ± 0.90	39.73^{a} ± 0.96	40.40^{a} ± 0.53	40.37^{a} ± 0.35	41.25^{a} ± 1.62	39.85^{a} ± 0.55	0.222
ADF	14.87^{a} ± 2.12	10.83^{a} ± 2.17	21.92^{b} ± 1.18	23.12^{b} ± 1.24	24.96^{b} ± 4.24	21.54^{b} ± 1.44	0.001
ADL	6.60^{a} ± 0.94	8.78^{a} ± 1.76	9.59^{a} ± 1.85	16.73^{b} ± 1.29	11.82^{ab} ± 2.76	10.56^{a} ± 3.15	0.021
Hemicellulose	23.68^{bc} ± 2.82	28.90^{c} ± 3.13	18.48^{ab} ± 1.68	17.24^{a} ± 1.18	16.30^{a} ± 2.61	18.31^{ab} ± 1.80	0.003
Cellulose	8.26^{a} ± 2.92	2.05^{a} ± 0.44	12.33^{a} ± 2.33	6.39^{a} ± 2.19	13.14^{a} ± 6.87	10.98^{a} ± 2.89	0.129
Weight loss (%)	6.34^{a} ± 5.9	6.36^{a} ± 3.91	6.29^{a} ± 2.35	6.65^{a} ± 1.96	6.45^{a} ± 6.30	9.88^{b} ± 3.8	0.016
Ca (mg/100g)	269.97^{a} ± 3.43	271.69^{a} ± 3.89	276.85^{a} ± 1.14	290.92^{b} ± 1.60	303.29^{c} ± 3.20	308.78^{c} ± 2.51	0.000
Fe (mg/100g)	0.49^{a} ± 0.02	0.79^{cd} ± 0.03	1.04^{e} ± 0.03	0.66^{b} ± 0.02	0.73^{bc} ± 0.03	0.84^{c} ± 0.02	0.000
Mg (mg/100g)	77.66^{a} ± 3.04	96.65^{b} ± 0.86	121.11^{cd} ± 4.87	123.55^{d} ± 4.86	115.28^{c} ± 1.32	117.27^{b} ± 0.93	0.000
Na (mg/100g)	4.41^{a} ± 0.30	7.89^{b} ± 0.18	7.65^{b} ± 0.47	7.54^{b} ± 0.30	7.43^{b} ± 0.04	7.52^{b} ± 0.12	0.000
K (mg/100g)	273.39^{a} 14.25	318.02^{b} 13.00	345.39^{c} 3.00	367.53^{d} 4.25	355.51^{cd} 3.75	359.66^{cd} 2.75	0.000
Cu (mg/100g)	0.37^{a} ± 0.05	0.49^{a} 0.06	0.66^{a} ± 0.11	0.77^{a} ± 0.10	0.57^{a} ± 0.05	0.55^{a} ± 0.25	0.213

Values for the same variable with different letters are significantly different. DM: Dry matter; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; ADL: Acid detergent lignin

The yeasts (Fig.3) multiplied for the first three days. During this period, they consumed the residual oxygen in the mixture. After that, their number decreased significantly from 11 to 6 for all treatments. After 15 days of silage, the lowest number of yeasts was found in the 10% treatment. The aerobic mesophilic bacteria multiplied less and above all in the treatment with 10% molasses (Fig.4). Total coliforms were detected in the 0%, 2%, and 4% treatments but disappeared after 10 days for the 2% and 4% treatments and 15 days for the molasses-free treatment (Fig.5). These results can be explained by the fact that sugars have a high capacity to bind water molecules, which produces a high osmotic pressure leading to the destruction of bacteria (Capozzi et al. 2009). Weise (1967) applied food-grade sugar to 10 kg/t of grass silage and reported that this stimulated LAB, Clostridia, and yeast. The author also reported that yeast was encouraged in sugar-

treated silage when air could infiltrate the silo. Furthermore, it should be noted that no proliferation of *E.coli*, fecal coliforms, *Staphylococci* and *Salmonella*, was detected throughout the silage process.

Table 4 Statistical significance of the mean effects

Parameters	Molasses level	Silage	Interaction (M*S)
pH	**	**	NS
DM	*	*	NS
Ash	NS	**	NS
Protein	**	**	**
Total sugar	**	**	**
Reducing sugar	**	**	**
NDF	**	**	**
ADF	**	**	**
ADL	*	**	NS
Cellulose	*	NS	NS
Hemicellulose	**	NS	NS
Ca	**	**	**
Fe	**	**	**
Mg	**	**	*
Na	**	**	**
K	**	**	*
Cu	**	**	NS

*: Significantly different at P < 0.05, **: Significantly different at P < 0.01, NS: Not significant

Aerobic stability test

Silage aerobic stability is important because silage is exposed to air during storage and feeding. The extent of air penetration into the silage during storage depends on its compaction and how the silo is sealed (Muck et al. 2003). The main spoilage microorganisms in the silage were aerobic yeasts (Fig.7), the growth of which was substantial after opening the silage bags. These yeasts can use both sugars and lactic acid. Lactic acid and residual sugars are the main energy source for microorganisms involved in silage spoilage (McDonald et al. 1991)**.**Yeasts play a major role in the aerobic deterioration of silage using lactate (Woolford 1990). Furthermore, yeast counts in silage can be useful because, as noted above, high numbers of yeasts in silage are generally associated with high concentrations of ethanol, and their numbers are often inversely related to the aerobic stability of the silage. Although they are relatively acid-tolerant and can utilize lactate present in silage when exposed to air, they are primarily involved in the aerobic deterioration of silage, which is accompanied by chemical changes, increased temperature, and loss of DM (Woolford 1990; Muck and Pitt 1993; Bolsen et al. 1996).

The metabolism of lactic and acetic acid by aerobic microorganisms leads to increased pH, which we found correlated with an increase in temperature (Figs.6, 8). However, after the active phase of fermentation ended, temperatures in the heart of the silo often decreased slowly to 25–30 °C. Small silos (including bag silos and large bales) need to cool down more quickly than large silos (Kung et al. 2018). The pH of the 10% treatment increased faster than that of the other treatments, while the 2% variant had the lowest pH value. The pH reached maximum values for all treatments at the end of aerobic exposure, which may be due to the decrease in lactic acid content with aerobic exposure time. The pH is an indicator of silage deterioration because the yeasts consume lactic acid during aerobic exposure, and the silage becomes favorable for the growth of other undesirable microorganisms such as molds and bacteria. Basso et al. (2012) and Hara and Ohyama (1978) also reported that the lactic acid content would decrease as the pH increased if the silage deteriorated.

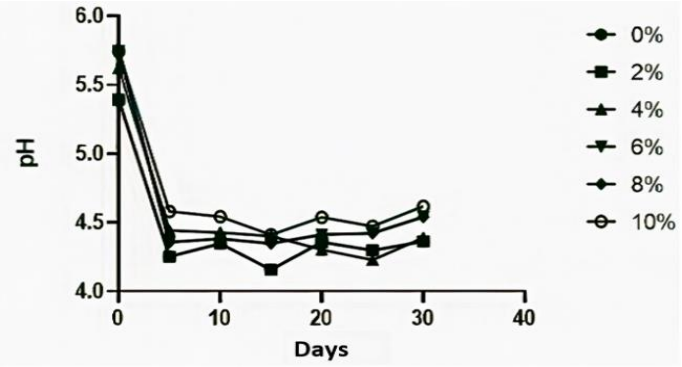

Fig. 1 Change in the pH value during the ensiling of cactus fruits with straw, wheat bran and different percentage of molasses

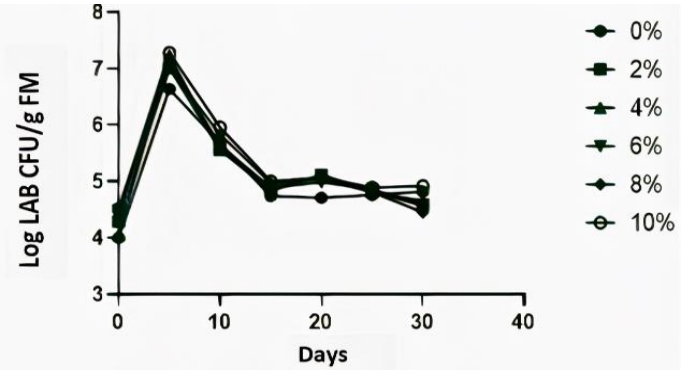

Fig. 2 Change in the development of lactic acid bacteria during the ensiling of cactus fruits with straw, wheat bran and different percentage of molasses

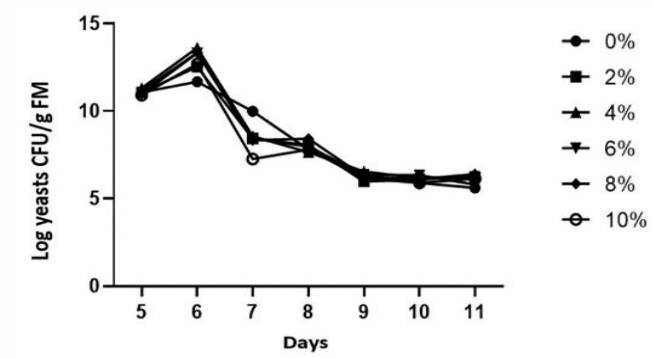

Fig. 3 Change in yeast development during the ensiling of cactus fruits with straw, wheat bran, and different percentages of molasses

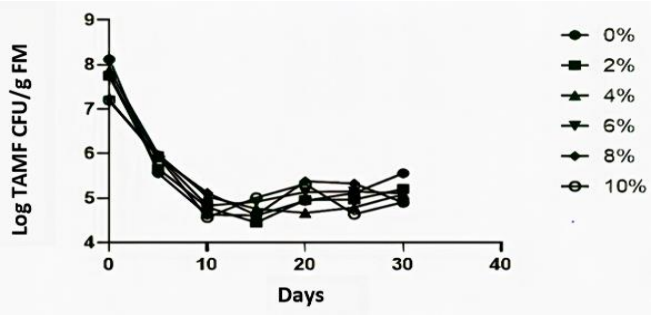

Fig. 4 Change in development of the total aerobic mesophilic flora during the ensiling of cactus fruits with straw, wheat bran, and different percentages of molasses

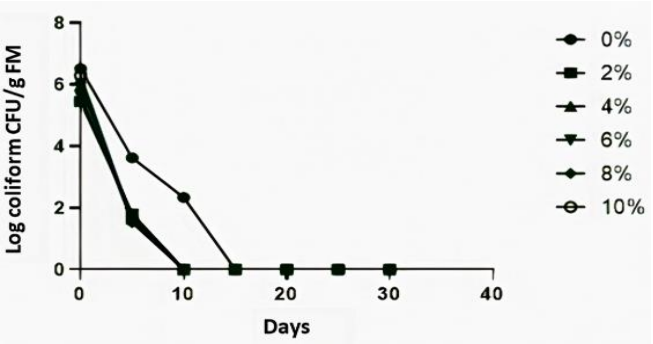

Fig. 5 Change in coliform bacteria during the ensiling of cactus fruits with straw, wheat bran, and different percentages of molasses

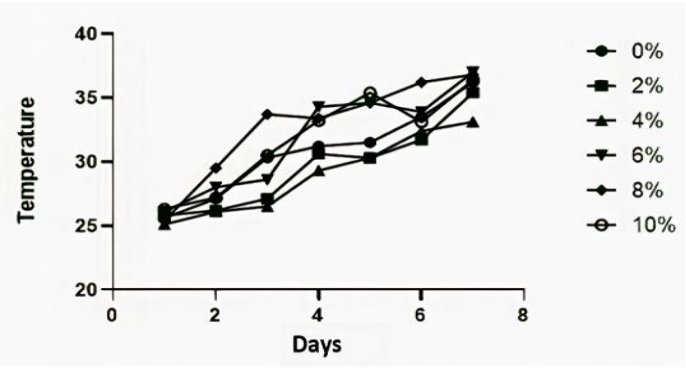

Fig. 6 Temperature during the aerobic stability test

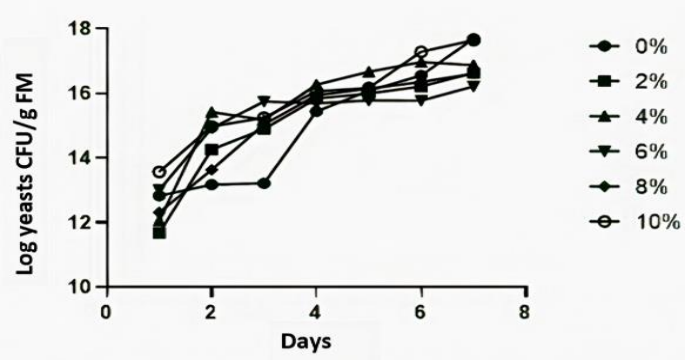

Fig. 7 Changes in yeast (log10) during the aerobic stability test

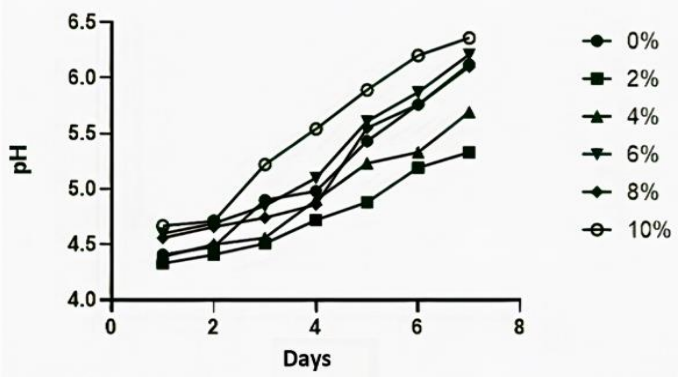

Fig. 8 pH during the aerobic stability test

Conclusion

Cactus mixtures can be preserved using a standard silage method, with or without molasses. The addition of molasses improves the fermentation and preservation process, limiting the silage losses. Furthermore, silage products are enhanced by reducing the growth of undesirable microorganisms. Therefore, molasses can be used effectively in *Opuntia ficus-indica* fruit silage. Our result suggest the addition of 10% beet molasse for optimum silage conservation. Our finding provide information for farmers to improve animal feed and denote an economic advantage considering the low cost of this silage production. Notwithstanding, the protein content levels remained very low, implying the necessity of protein enrichment. Ultimately, further work, especially on animal responses to specific silage, is required to confirm the reported nutritional characteristics of this study.

Compliance with ethical standards

Conflict of interest The authors declare that there are no conflicts of interest associated with this study.

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