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Effects of Sour Yogurt as an Alternative Additive in Second Crop Corn Silage

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Abstract: In this study, we evaluated the influence of sour yogurt as a natural microbial inoculant in second-crop corn silages. For this purpose, two trials with different dilution rates were conducted. In Trial I, the groups 10 g sour yogurt + 5 g distilled water (SY10-2), 20 g sour yogurt + 10 g distilled water (SY20-2), 30 g sour yogurt + 15 g distilled water (SY30-2), 40 g sour yogurt + 20 g distilled water (SY40-2), 50 g sour yogurt + 25 g distilled water (SY50-2) and no additives were added to the control (CON) group. The groups in Trial II, 10 g sour yogurt + 10 g distilled water (SY10-1), 20 g sour yogurt + 20 g distilled water (SY20-1), 30 g sour yogurt + 30 g distilled water (SY30-1), 40 g sour yogurt + 40 g distilled water (SY40-1), 50 g sour yogurt + 50 g distilled water (SY50-1) and 10 g of distilled water were added to the control (WCON) group. For the silages opened on the 90th day of ensiling, the highest lactic acid content was determined in the SY20-2 and SY20-1 groups ($p < 0.05$). The lowest amount of ammonia nitrogen was in the SY30-2 group ($p < 0.05$). In the aerobic period, the SY10-2 and SY20-2 groups remained more stable than the others. As a result, the SY20-2, SY30-2, SY20-1, and SY30-1 groups improved the fermentation quality of corn silages, but the effect on aerobic stability was not significant and was similarly found with the homofermentative bacterial inoculants.

Keywords: sour yogurt; corn; silage; fermentation quality; in vitro digestibility; animal nutrition

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1. Introduction

Corn (*Zea mays* L.) is the most commonly used plant in silage production worldwide [1]. Corn is an ideal silage crop with a relatively high dry matter (DM) content, low buffer capacity, and sufficient water-soluble carbohydrates for lactic acid (LA) fermentation [2].

In Turkey, in the 2020/2021 season, there was a shrinkage in the first crop corn cultivation area, but the total production increased due to the growth in the second crop corn cultivation area. This year (2021/2022), the second crop corn production is expected to increase [3]. It is not easy to harvest at optimum dry matter (30–35%) because the harvest time for silage corn, when planted as a second crop, is in October, when the precipitation increases [4]. Since the corn plant is not suitable for withering, it is imperative to add additives to the low DM corn to be silage [5].

Homofermentative lactic acid bacteria (LAB), one of the bacterial silage inoculants, is very effective in minimizing carbon dioxide losses at the beginning of silage fermentation. They have been developed to grow rapidly in silage, lower the pH and dominate fermentation [6]. In most studies [7,8], it has been reported that homofermentative inoculants are successful in lowering the pH and shifting fermentation to lactate and can reduce fermentation losses. The effects of homofermentative LAB inoculants on aerobic stability were variable. Ranjit and Kung [9] reported that homofermentative LAB inoculants improved the aerobic stability of silages, while Hu et al. [10] reported that it did not affect it, and Muck [7] reported that it reduced it.

It is thought that by using food industry by-products in animal nutrition, this product will reduce pollution and damage to the environment, as well as provide added value to animal husbandry [11,12]. Yogurt is a fermented dairy product produced by *Lactobacillus bulgaricus* and *Streptococcus thermophilus* bacteria [13]. These bacteria convert six-carbon sugars into lactic acid and various carbonyl compounds homofermentatively through pyruvate metabolism [14]. The main bacteria in sour yogurt are *L. bulgaricus* and *S. thermophilus* [15]. Kiani et al. [16] reported that the addition of 5% sour yogurt on a dry matter basis significantly decreased the pH, crude ash (CA), and ammonia nitrogen (NH_3-N) values of corn silage and increased the Flieg score, crude protein (CP) and total nitrogen. Researchers also stated that the effects of sour yogurt addition on corn silage quality should be investigated together with detailed chemical properties and animal production. Sour yogurt is a waste product that frequently occurs in homes, markets, and small and large-scale livestock enterprises in Turkey. Sour yogurt can be an alternative to bacterial inoculants because it contains homofermentative bacteria, is easily available, has no cost, and is practical. Sour yogurt can be easily used in livestock enterprises to reduce silage costs and improve silage quality; thus, it can be recycled back into the economy. However, due to the low DM content of second-crop maize, the dilution rate and dose should be determined as well.

This study aims to reveal the potential of being an alternative to bacterial inoculants by examining the effects of adding sour yogurt as a natural source of lactic acid bacteria to the second crop of corn harvested in October on the fermentation quality, aerobic stability, and in vitro digestibility.

2. Materials and Methods

2.1. Experimental Design and Ensiling Process

The research material was the second product, corn (Pioneer 32K61) and sour yogurt. Corn (41.12° N and 27.6° S, Hayrabolu, Tekirdağ, Turkey) was cut in the dough stage in October and chopped to 1.5–2 cm. Before ensiling, fresh corn (FC) contained 23.70% dry matter (DM), 6.2% CA of DM, 7.8% CP of DM, 117.9 g/kg DM of water-soluble carbohydrate (WSC), 280.1 mEq NaOH kg/DM of buffer capacity (Bc), 2.07 log CFU/g of *Lactobacilli*, 2.0 log CFU/g of *Lactococci*, 1.91 log CFU/g of yeast, and 1.93 CFU/g of enterobacter and no mold, with a pH of 5.6.Since the expiration date was approaching, the sour-tasting yogurt was bought from the market. Sour yogurt (natural) had 12.99% DM, 0.97% CA, 5.48% CP, 90.29 g/kg DM of WSC, 115.95 g/kgDM of LA, 3.5 of pH, 4.54 log CFU/gof *Lactobacilli*, 2.0 log CFU/g of *Lactococci*, and 5.0 log CFU/g of yeast, with no mold before ensiling.

In Trial I, the groups 10 g sour yogurt + 5 g distilled water (SY10-2), 20 g sour yogurt + 10 g distilled water (SY20-2), 30 g sour yogurt + 15 g distilled water (SY30-2), 40 g sour yogurt + 20 g distilled water (SY40-2), 50 g sour yogurt + 25 g distilled water (SY50-2) and no additives were added to the control (CON) group. The groups in Trial II of10 g sour yogurt + 10 g distilled water (SY10-1), 20 g sour yogurt + 20 g distilled water (SY20-1), 30 g sour yogurt + 30 g distilled water (SY30-1), 40 g sour yogurt + 40 g distilled water (SY40-1), 50 g sour yogurt + 50 g distilled water (SY50-1) and 10 g of distilled water were added to the control (WCON) group. The additives were added to 1 kg of fresh corn. Since the DM content of corn is low at 23.70%, the lowest level of distilled water was added to the WCON group so as not to increase the silo water output [2].

The silage was made in a laboratory-scale fermentation system: approximately 500 g of corn was weighed, and after the corn was placed in oxygen-barrier polyethylene bags, they were vacuumed (CAS CVP−260PD) for 25 s at a vacuum level of 0.1 mPa [17]. It has been reported that high-quality silages are obtained, even at the lowest vacuum level [17,18]. A total of 48 packages, 4 for each group, were left to ferment for 90 days under laboratory conditions (10–20 °C).

2.2. Physically and Chemical Analysis

Three different observers scored the silages on the day they were opened (90th day) in terms of their color, odor and structure (Deutsche Landwirtschafts Gesellschaft: DLG) [19]. The evaluation, according to DLG, is 16–20: excellent; 10–15: moderate; 5–9: medium; 0–4: poorly [19]. Immediately after the bags were opened, the subsamples (20 g) were blended with 180 mL of distilled water for 1 h. The extracts were filtered through four layers of cheesecloth. The pH of the silages was determined with a digital pH meter, the buffer capacity was determined according to Playne and McDonald [20], and the LA was by the spectrophotometric method [21]. The ammonia nitrogen and WSC contents were determined according to Anonymous [22]. Silage volatile fatty acids (acetic, butyric, propionic acid) and ethanol were analyzed in silage liquid. Silage liquid was taken at 10 mL and placed into tubes (15 mL). Then, 1.0 mL of 25% phosphoric acid was added to the silage liquid [23]. The silage liquid was centrifuged at 35,060× *g*. Gas chromatography (GC, Agilent 6890N) was used in the chemical analysis of the volatile fatty acids and ethanol. The Stabilwax-DA capillary column (30.0 m, 0.25 mm ID, 0.25 µm) and helium carrier gas (22 mL/min linear velocity, 25:1 split ratio, and 230 °C temperature) were used in the GC system.

A five-day aerobic stability test was carried out on the samples developed by Ashbell et al. [24]. The Flieg score was calculated from the silage's dry matter and pH values according to the formula below [19].

$$\text{Flieg score} = 220 + (2 \times \%\text{DM} - 15) - 40 \times \text{pH}.$$

According to this index, silage was considered "poor" when it had a score of <20; to be "low" with a score between 21 and 40; to be "medium" with a score between 41 and 60; to be "good" quality with a score between 61 and 80; to be "excellent" when it had a score between 81 and 100 [19,25].

2.3. Microbial Populations

Lactobacilli, *lactococci*, yeast and mold analyses were determined by the method developed by Seale et al. [26]. MRS agar (de Man Rogosa and Sharpe agar, Merck, Darmstadt, Germany) was used to detect *lactobacilli*. *Lactococci* were determined on M17 agar (Merck) [27]. In the enumeration of the yeast, malt extract agar was used, and violet red bile agar was used for enterobacter. The plates were incubated for 3 days at 30 °C. The *Lactobacilli*, *lactococci*, mold and yeast numbers of the silages were converted into logarithmic colony-forming units (CFU/g).

2.4. Nutrient Analysis and In Vitro Digestibility

The DM was determined by drying the samples at 105 °C for 16 h. The organic matter (OM), CP, and CA contents of feed samples were determined by AOAC [28]. The neutral detergent fiber (NDF), acid detergent insoluble fiber (ADF), and acid detergent insoluble lignin (ADL) contents were determined according to the methods reported by Van Soest et al. [29].

Pepsin-cellulase digestibility was determined according to a modification of De Boever et al.'s method [30]. In the technique [31], pre-treatment with the pepsin-hydrochloric acid solution followed an incubation in water at 80 °C for 45 min before the cellulase treatment (Onozuka R 10 from *Trichoderma viride*, Merck). The solubility of the organic matter in cellulase (ELOS), the cellulase digestibility of the organic matter (DOM), and the insoluble organic matter in cellulase (EULOS) were derived as follows:

$$\text{ELOS } (\%) = \text{DM-CA} - \text{G}$$
$$\text{G } (\%) = \text{Loss upon ashing}$$
$$\text{DOM } (\%) = (\text{ELOS} \times 10^2/100 - \text{CA}\%)$$
$$\text{EULOS } (\text{g/kg}) = 1000 - \text{CA } (\text{g/kg DM}) - (\text{ELOS}\% \times 10)$$

The following equation, reported by Weissbach et al., was used to determine the metabolic energy contents [32]. Once obtained, the ME contents were translated into kilocalories.

$$\text{ME (MJ/kg DM)} = 13.98 - 0.0147 \times \text{CA} - 0.0102 \times \text{EULOS} - 0.00000254 \times \text{EULOS}^2 + 0.00234 \times \text{CP}$$

With CA, EULOS, CP in g/kg DM.

2.5. Statistical Analyses

The statistical analyses were performed using the SPSS software v.18 suite [33]. The effects of different treatments were evaluated using a one-way analysis of variance with Duncan's multiple range tests. The relationships between the LAB and nutrient contents and some silage quality parameters were determined by Pearson's correlation analysis. The smell, structure, color and DLG points of silages were analyzed via nonparametric Kruskal–Wallis tests. Differences were significant when $p < 0.01$ and $p < 0.05$ [34].

3. Results

3.1. Trial I

According to the evaluation, while adding sour yogurt to the corn affected the odor positively, it did not harm the structure and positively affected the color (Table 1). Especially in the SY20-2 and SY30-2 groups, the smell was more pleasant; the color was more vivid and green, and the structure of the leaf-stem integrity remained as fresh as the first day. The highest Flieg score was determined in the SY30-2 group (110.18) ($p < 0.05$). In addition, the SY20-2, SY10-2 and SY40-2 groups were determined by Flieg scores, respectively, 109.39, 109.31 and 108.18 ($p < 0.05$).

Table 1. The effects of different yogurt levels on silage qualities ($n = 4$).

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
Smell	8.0	14.0	14.0	14.0	14.0	14.0	0.47	<0.001
Structure	4.0	4.0	4.0	4.0	4.0	4.0	0.00	1.000
Colors	1.0	2.0	2.0	2.0	1.0	1.0	0.10	<0.001
DLG point	13	20	20	20	19	19	0.52	<0.001
Quality	Moderate	Excellent	Excellent	Excellent	Excellent	Excellent		
Flieg Point	103.67 ^{b}	109.31 ^{a}	109.39 ^{a}	110.18 ^{a}	108.18 ^{a}	105.34 ^{b}	0.50	<0.001
Quality	Excellent	Excellent	Excellent	Excellent	Excellent	Excellent		

CON, control no additives; SY10-2, 10 g sour yogurt + 5 g distilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; SEM, standard error of the mean. ^{a,b}, Means with different letters in the same line are statistically significant ($p < 0.05$).

The crude nutrient and cell wall contents of the silages are given in Table 2. The addition of sour yogurt at different rates significantly affected the chemical composition of the silages; while the amount of CP (from 7.93% to 8.68%) and CA (from 5.96% to 6.25%) increased depending on the dose, and NDF (63.24%) and ADF (28.36%) of the cell wall components were found to be the highest in the PY50-2 group ($p < 0.05$).The NDF amount of the CON group was found to be similar to the SY10-2 and SY30-2 groups ($p > 0.05$).The ADL contents were found to be between 3.36–3.94% in all groups. The addition of sour yogurt significantly ($p < 0.05$) increased the DM of the SY30-2 group (Table 3) compared to the CON group. The DM amounts of the SY10-2 and SY20-2 groups were 22.36% and 22.39%, respectively. The DM amounts of the SY10-2, SY20-2 and SY30-2 groups are significant compared to the CON and other yogurt groups ($p < 0.05$). The silage pH was determined between 3.51–3.63 in all groups ($p < 0.05$). The WSC contents were also lower in the yogurt groups than in the CON group ($p < 0.05$). The highest LA content was determined in the SY20-2 group ($p < 0.05$) and the lowest in the CON. The increase in the amount of LA in the yogurt-added groups, according to the CON, was found to be statistically significant ($p < 0.05$). Acetic acid (AA) and butyric acid contents decreased inversely with the increase in yogurt levels ($p < 0.05$). Propionic acid, ethanol and LA/AA were found to increase in parallel with increasing yogurt levels ($p < 0.05$). The addition of different levels of sour yogurt caused a decrease ($p < 0.05$) in the amount of NH_3-N in all groups compared to the CON. This decrease was found to be statistically significant ($p < 0.05$). The NH_3-N amounts were determined in the SY10-2, SY20-2, SY30-2, SY40-2 and SY50-2 groups, respectively, in the 45.98, 38.13, 38.56, 35.70, 36.94 and 39.61 g/kgTN (Table 3).

Table 2. Chemical compositions of the corn silages (% in DM).

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
OM	94.58 ^{a}	94.05 ^{b,c}	93.88 ^{c,d}	93.94 ^{c,d}	94.22 ^{b}	93.75 ^{d}	0.07	<0.001
CP	7.85 ^{d}	7.93 ^{c,d}	7.97 ^{b,c,d}	8.16 ^{b,c}	8.22 ^{b}	8.68 ^{a}	0.07	<0.001
CA	5.42 ^{d}	5.96 ^{b,c}	6.12 ^{a,b}	6.06 ^{a,b}	5.78 ^{c}	6.25 ^{a}	0.07	<0.001
NDF	56.66 ^{d}	56.34 ^{d}	58.70 ^{c}	56.47 ^{d}	60.06 ^{b}	63.24 ^{a}	0.61	<0.001
ADF	24.57 ^{e}	27.77 ^{b}	27.84 ^{b}	27.30 ^{c}	26.76 ^{d}	28.36 ^{a}	0.30	<0.001
ADL	3.93 ^{a}	3.68 ^{b}	3.94 ^{a}	3.78 ^{a,b}	3.28 ^{c}	3.36 ^{c}	0.07	<0.001

CON, control no additives; SY10-2, 10 g sour yogurt + 5 g distilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; OM, organic matter; CP, crude protein; CA, crude ash; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin; SEM, standard error of means. $^{a–e}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

Table 3. Fermentation quality of corn silages.

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
DM,%	22.00 ^{c}	22.36 ^{b}	22.39 ^{b}	22.79 ^{a}	21.92 ^{c}	20.44 ^{d}	0.18	<0.001
pH	3.63 ^{a}	3.51 ^{b}	3.51 ^{b}	3.51 ^{b}	3.52 ^{b}	3.51 ^{b}	0.01	<0.001
WSC, g/kgDM	18.65 ^{a}	16.48 ^{b}	15.53 ^{d}	15.46 ^{d}	16.03 ^{c}	16.49 ^{b}	0.26	<0.001
LA, g/kgDM	69.89 ^{e}	71.07 ^{b,c}	71.99 ^{a}	71.30 ^{b}	70.91 ^{c}	70.39 ^{d}	0.16	<0.001
AA, g/kg DM	30.85 ^{a}	30.52 ^{a}	28.05 ^{b}	26.76 ^{c}	25.31 ^{d}	24.13 ^{e}	0.61	<0.001
BA, g/kg DM	1.55 ^{a}	1.50 ^{a}	1.38 ^{c}	1.25 ^{d}	1.25 ^{d}	1.04 ^{e}	0.04	<0.001
PA, g/kgDM	1.88 ^{e}	2.11 ^{d}	2.17 ^{c}	2.36 ^{b}	2.51 ^{a}	2.47 ^{a}	0.05	<0.001
Ethanol, g/kgDM	2.03 ^{e}	2.15 ^{d}	2.31 ^{c}	2.53 ^{a,b}	2.56 ^{a}	2.50 ^{b}	0.05	<0.001
LA/AA	2.27 ^{e}	2.33 ^{e}	2.57 ^{d}	2.67 ^{c}	2.80 ^{b}	2.92 ^{a}	0.06	<0.001
NH_3-N, g/kgTN	45.98 ^{a}	38.13 ^{d}	38.56 ^{c}	35.70 ^{f}	36.94 ^{e}	39.61 ^{b}	0.79	<0.001
DM loss,%	1.24 ^{c}	1.55 ^{a,b}	1.49 ^{b}	1.59 ^{a,b}	1.64 ^{a,b}	1.67 ^{a}	0.04	0.001

CON, control no additives; SY10-2, 10 g sour yogurt + 5 g distilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; WSC, water-soluble carbohydrates; LA, lactic acid; AA, acetic acid; BA, butyric acid; PA, propionic acid; LA/AA, the ratio of lactic acid and acetic acid; NH_3-N, ammonia nitrogen; TN, total nitrogen; DM loss, dry matter loss, SEM, standard error of the mean. $^{a–f}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

As seen in Table 4, it was found that the *Lactobacilli* counts increased significantly ($p < 0.05$) in the groups with added sour yogurt compared to the CON, while the *Lactococci* counts increased only in the SY10-2 (5.27 log10 CFU/g) and SY20-2 (5.23 log10 CFU/g) groups ($p < 0.05$). It was determined that the yeast and enterobacter counts decreased in yogurt groups compared to the CON ($p < 0.05$). In the study, no mold growth was observed in any group.

Table 4. Microbiological analysis results of corn silages, log10 CFU/g.

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
Lactobacilli	4.14 ^{e}	5.23 ^{d}	5.50 ^{c}	5.61 ^{b,c}	5.74 ^{b}	5.93 ^{a}	0.14	<0.001
Lactococci	3.31 ^{b}	5.27 ^{a}	5.23 ^{a}	3.51 ^{b}	2.94 ^{c}	2.82 ^{c}	0.25	<0.001
Yeast	3.97 ^{a}	3.54 ^{b}	3.43 ^{b}	3.51 ^{b}	3.42 ^{b}	3.41 ^{b}	0.05	0.001
Mold	ND	ND	ND	ND	ND	ND	-	-
Enterobacter	2.92 ^{a}	1.2 ^{b}	ND^{c}	1.11 ^{b}	1.26 ^{b}	1.10 ^{b}	0.22	<0.001

CON, control no additives; SY10-2, 10 g sour yogurt + 5 gdistilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; ND, not detected; CFU, colony-forming units; SEM, standard error of the mean. $^{a–e}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

When the aerobic stability test results (Table 5) were examined, it was observed that the addition of a low amount of yogurt reduced the yeast counts and CO_2 output, but it could not prevent yeast growth ($p < 0.05$) compared to the CON, and mold growth did not exist. The DM contents of the CON, SY10-2, SY20-2, SY30-2, SY40-2 and SY50-2 groups were found to be 21.28, 21.55, 20.65, 19.93, 19.02 and 18.63%, respectively.

Table 5. Aerobic stability test results of corn silages.

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
DM,%	21.28 ^{a}	21.55 ^{a}	20.65 ^{b}	19.93 ^{c}	19.02 ^{d}	18.63 ^{e}	0.27	<0.001
pH	6.00 ^{d}	6.67 ^{c}	6.77 ^{b,c}	6.87 ^{a,b}	6.90 ^{a,b}	7.00 ^{a}	0.08	<0.001
CO_2 g/kg DM	118.65 ^{d}	114.50 ^{f}	115.58 ^{e}	120.21 ^{c}	122.17 ^{b}	124.09 ^{a}	0.83	<0.001
Yeast, log CFU/g	6.09 ^{c}	5.44 ^{e}	5.76 ^{d}	6.36 ^{b}	6.58 ^{b}	6.81 ^{a}	0.11	<0.001
Mold, log CFU/g	ND	ND	ND	ND	ND	ND	-	-

CON, control no additives; SY10-2, 10 g sour yogurt + 5 gdistilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; ND, not detected; CFU, colony-forming units; SEM, standard error of the mean. $^{a–f}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

As seen in Table 6, the highest ELOS and DOM in corn silages were 65.30% and 69.52% in the SY30-2 group, respectively ($p < 0.05$). In comparison, the lowest amount of EULOS was determined in the SY30-2 group. The in vitro ME contents calculated from EULOS were the highest (2525.6 kcal/kg DM) in this group ($p < 0.05$).

Table 6. In vitro digestibility and ME contents of corn silages (in DM).

Item	CON	SY10-2	SY20-2	SY30-2	SY40-2	SY50-2	SEM	*p*-Value
ELOS,%	63.87 ^{b,c}	63.68 ^{c}	63.44 ^{c}	65.30 ^{a}	62.82 ^{d}	64.23 ^{b}	0.19	<0.001
EULOS, g/kg	307.11 ^{b}	303.64 ^{b}	304.38 ^{b}	286.36 ^{d}	313.96 ^{a}	295.18 ^{c}	2.19	<0.001
DOM, %	67.53 ^{c}	67.71 ^{c}	67.58 ^{c}	69.52 ^{a}	66.68 ^{d}	68.51 ^{b}	0.22	<0.001
ME, kcal/kg	2503.2 ^{b}	2492.1 ^{b,c}	2484.9 ^{c}	2525.6 ^{a}	2478.6 ^{c}	2503.5 ^{b}	3.98	<0.001

CON, control no additives; SY10-2, 10 g sour yogurt + 5 g distilled water; SY20-2, 20 g sour yogurt + 10 g distilled water; SY30-2, 30 g sour yogurt + 15 g distilled water; SY40-2, 40 g sour yogurt + 20 g distilled water; SY50-2, 50 g sour yogurt + 25 g distilled water; ELOS, solubility of the organic matter in cellulase; EULOS, insoluble organic matter in cellulase; DOM, digestible organic matter; SEM, standard error of the mean. $^{a–d}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

3.2. Trial II

When Table 7 is examined, especially in the SY20-1 and SY30-1 groups, the smell is more pleasant, the color is more vivid, and the green has preserved its freshness of the first day. The Flieg scores for the CON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 106.7, 107.2, 108.4, 108.7, 107.3 and 107.2, respectively. The difference between the groups was statistically insignificant ($p > 0.05$).

Table 7. The effects of different yogurt levels on silage qualities ($n = 4$).

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
Smell	8.0	14.0	14.0	14.0	14.0	14.0	0.47	<0.001
Structure	4.0	4.0	4.0	4.0	4.0	4.0	0.000	1.000
Colors	1.0	2.0	2.0	2.0	1.0	1.0	0.10	<0.001
DLG point	13	20	20	20	19	19	0.52	<0.001
Quality	Moderate	Excellent	Excellent	Excellent	Excellent	Excellent	-	-
Flieg Point	106.7	107.2	108.4	108.7	107.3	107.2	0.39	0.694
Quality	Excellent	Excellent	Excellent	Excellent	Excellent	Excellent	-	-

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water; SEM, standard error of the mean.

In this study (Table 8), adding diluted sour yogurt to corn at different rates and one-to-one caused an increase in the CP contents ($p < 0.05$) especially. It was determined that the CP contents of the SY20-1, SY30-1, SY40-1 and SY50-1 groups increased significantly ($p < 0.05$) compared to the CON and SY10-1 groups. Among the cell wall components, the NDF and ADF contents were found to be the lowest in the SY30-1 group (NDF: 54.94%, ADF: 23.17%) ($p < 0.05$). The NDF and ADF contents were found to be similar in the SY20-1 and SY40-1 groups. The ADL contents were found to be between 3.29–3.96% in all groups.

Table 8. Chemical compositions of the corn silages (% in DM).

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
OM	94.51 ^{a}	94.08 ^{d}	94.37 ^{a,b,c}	94.23 ^{c,d}	94.28 ^{b,c}	94.45 ^{a,b}	0.04	0.003
CP	7.66 ^{c}	7.75 ^{c}	8.09 ^{b}	7.85 ^{c}	8.45 ^{a}	8.52 ^{a}	0.08	<0.001
CA	5.50 ^{d}	5.92 ^{a}	5.63 ^{b,c,d}	5.77 ^{a,b}	5.72 ^{b,c}	5.55 ^{c,d}	0.04	0.003
NDF	58.77 ^{b}	58.08 ^{b}	56.83 ^{c}	54.94 ^{d}	56.43 ^{c}	61.21 ^{a}	0.49	<0.001
ADF	26.59 ^{b}	27.61 ^{a}	26.88 ^{b}	23.17 ^{d}	26.56 ^{b}	25.96 ^{c}	0.34	<0.001
ADL	3.58 ^{b,c}	3.96 ^{a}	3.67 ^{b}	3.34 ^{c}	3.37 ^{b,c}	3.29 ^{c}	0.06	0.002

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water; OM, organic matter; CP, crude protein; CA, crude ash; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin; SEM, standard error of means. $^{a–d}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

The transaction positively affected the DM (Table 9) of the PY30-1 group (22.66%) and reduced the DM loss (1.58%). The silage pH was not affected by the addition of sour yogurt, and the pH was determined between 3.51–3.54 in all groups ($p > 0.05$). The WSC contents were also lower in the yogurt groups compared to the WCON group (21.64 g/kg DM). The highest LA content was determined in the SY30-1 group ($p < 0.05$), and the lowest was in the WCON group. The acetic acid and butyric acid contents of silages decreased inversely with the increase in yogurt levels. The decrease in the amount of acetic acid is statistically significant ($p < 0.05$). Propionic acid, ethanol and LA/AA were found to increase in parallel with the increasing yogurt level ($p < 0.05$). The ethanol contents in the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 1.96, 2.04, 2.11, 2.12, 2.23 and 2.18 g/kg DM, respectively. Sour yogurt, added at different levels, caused a decrease ($p < 0.05$) in the amount of NH_3-N compared to the WCON group (46.40 g/kg DM).

Table 9. Fermentation quality of corn silages.

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
DM,%	21.51 ^{c}	21.77 ^{b,c}	21.88 ^{b}	22.66 ^{a}	21.61 ^{b,c}	21.56 ^{c}	0.10	<0.001
pH	3.53	3.53	3.51	3.54	3.52	3.52	0.01	0.963
WSC, g/kg DM	21.64 ^{a}	16.48 ^{b}	15.53 ^{d}	15.35 ^{d}	16.03 ^{c}	16.49 ^{b}	0.52	<0.001
LA, g/kg DM	67.29 ^{e}	79.42 ^{c}	80.71 ^{b}	84.58 ^{a}	79.59 ^{c}	77.89 ^{d}	1.29	<0.001
AA, g/kg DM	28.68 ^{a}	27.28 ^{b}	25.51 ^{c}	23.78 ^{d}	23.46 ^{d}	22.97 ^{d}	0.52	<0.001
BA, g/kg DM	1.44 ^{a}	1.40 ^{a}	1.34 ^{b}	1.21 ^{c}	1.14 ^{d}	1.00 ^{e}	0.04	<0.001
PA, g/kg DM	1.96 ^{d}	2.04 ^{c}	2.11 ^{b}	2.12 ^{b}	2.23 ^{a}	2.18 ^{a}	0.02	<0.001
Ethanol, g/kg DM	1.98 ^{d}	2.11 ^{c}	2.32 ^{b}	2.41 ^{b}	2.54 ^{a}	2.54 ^{a}	0.52	<0.001
LA/AA	2.35 ^{e}	2.91 ^{d}	3.16 ^{c}	3.56 ^{a}	3.39 ^{b}	3.39 ^{b}	0.09	<0.001
NH_3-N, g/kg TN	46.40 ^{a}	42.49 ^{c}	40.04 ^{e}	40.86 ^{d}	42.75 ^{c}	43.20 ^{b}	0.49	<0.001
DM Loss,%	1.81 ^{a,b}	1.64 ^{c}	1.73 ^{a,b,c}	1.58 ^{c}	1.67 ^{b,c}	1.85 ^{a}	0.03	0.010

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water; WSC, water-soluble carbohydrates; LA, lactic acid; AA, acetic acid; BA, butyric acid; PA, propionic acid; LA/AA, the ratio of lactic acid and acetic acid;NH_3-N, ammonia nitrogen; TN, total nitrogen; DM loss, dry matter loss; SEM, standard error of the mean. $^{a–e}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

The microbiological analysis results of the silages are given in Table 10. With the addition of sour yogurt, the *Lactobacilli* and *Lactococci* counts increased significantly compared to the WCON ($p < 0.05$). The *Lactobacilli* counts in the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 4.24, 5.36, 5.35, 5.28, 5.26 and 4.78 log10 CFU/g, respectively, and the *Lactococci* counts in the groups were determined as 3.49, 4.69, 4.36, 4.23, 4.11 and 3.98 log10 CFU/g, respectively. It was determined that the yeast counts decreased in the SY10-1 (3.48 log10 CFU/g), SY20-1 (3.52 log10 CFU/g), and SY30-1 (3.46 log10 CFU/g) groups compared to the WCON (3.84 log10 CFU/g) group ($p < 0.05$), while the enterobacter counts were higher in the yogurt groups compared to the WCON group. In the study, it was observed that there was no mold growth in any group.

Table 10. Microbiological analysis results of corn silages, log10 CFU/g.

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
Lactobacilli	4.24 ^{c}	5.36 ^{a}	5.35 ^{a}	5.28 ^{a}	5.26 ^{a}	4.78 ^{b}	0.10	<0.001
Lactococci	3.49 ^{d}	4.69 ^{a}	4.36 ^{b}	4.23 ^{b,c}	4.11 ^{b,c}	3.98 ^{c}	0.09	<0.001
Yeast	3.84 ^{a}	3.48 ^{c}	3.52 ^{c}	3.46 ^{c}	3.70 ^{b}	3.75 ^{a,b}	0.04	<0.001
Mold	ND	ND	ND	ND	ND	ND	-	-
Enterobacter	1.49 ^{a}	2.35 ^{a}	2.16 ^{a,b}	2.00 ^{b}	2.14 ^{a,b}	2.39 ^{a}	0.08	0.01

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water; ND, not detected; CFU, colony-forming units; SEM, standard error of the mean. $^{a–d}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

When the aerobic stability results are examined in Table 11, it demonstrates that a low amount of yogurt addition (SY10-1, SY20-1, respectively, 5.90 and 5.57) reduced the pH and CO_2 output, but it could not prevent yeast growth ($p < 0.05$), and mold growth was not found in all groups. The DM contents in the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 20.64, 20.17, 21.16, 20.20, 19.77 and 19.89%, respectively. Additionally, the yeast counts in the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 6.51, 5.25, 5.33, 5.79, 5.82 and 5.83 log10 CFU/g, respectively.

Table 11. Aerobic stability test results of corn silages.

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
DM, %	20.64 ^{b}	20.17 ^{c,d}	21.16 ^{a}	20.20 ^{c}	19.77 ^{d}	19.89 ^{c,d}	0.12	<0.001
pH	6.63 ^{b}	5.90 ^{c}	5.57 ^{d}	6.60 ^{b}	6.80 ^{a}	6.87 ^{a}	0.12	<0.001
CO_2 g/kg DM	128.05 ^{b}	125.83 ^{c}	119.83 ^{e}	123.67 ^{d}	125.56 ^{c}	132.43 ^{a}	0.94	<0.001
Yeast, log CFU/g	6.51 ^{a}	5.25 ^{d}	5.33 ^{c}	5.79 ^{b}	5.82 ^{b}	5.83 ^{b}	0.09	<0.001
Mold, log CFU/g	ND	ND	ND	ND	ND	ND	-	-

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water;ND, Not detected; CFU, Colony-forming units; SEM, standard error of the mean. $^{a–e}$ Means with different letters in the same line are statistically significant ($p < 0.05$).

The in vitro digestibility and ME contents of the corn silages are given in Table 12. The ELOS contents of the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 63.11, 64.02, 65.48, 66.93, 66.70 and 66.58%, respectively. The increase in the amount of ELOS in the groups with yogurt added compared to the WCON group was statistically significant ($p < 0.05$). The digestibility of organic matter in the enzyme was found to be the highest in the SY30-1, SY40-1, and SY50-1 groups, respectively, at 71.03, 70.75, and 70.49% ($p < 0.05$). The EULOS contents of the WCON, SY10-1, SY20-1, SY30-1, SY40-1 and SY50-1 groups were determined as 313.95, 300.63, 288.91, 272.98, 275.77 and 278.22 g/kg DM, respectively. In addition, the in vitro ME contents that were calculated from the EULOS contents were found to be higher in the SY30-1 (2562.2 kcal/kg DM), SY40-1 (2561.4 kcal/kg DM), and SY50-1 (2561.6 kcal/kg DM) groups ($p < 0.05$).

Table 12. In vitro digestibility and ME contents of corn silages (in DM).

Item	WCON	SY10-1	SY20-1	SY30-1	SY40-1	SY50-1	SEM	*p*-Value
ELOS, %	63.11 [d]	64.02 [c]	65.48 [b]	66.93 [a]	66.70 [a]	66.58 [a]	0.36	<0.001
EULOS, g/kg	313.95 [a]	300.63 [b]	288.91 [c]	272.98 [e]	275.77 [d,e]	278.77 [d]	3.61	<0.001
DOM, %	66.78 [d]	68.05 [c]	69.39 [b]	71.03 [a]	70.75 [a]	70.49 [a]	0.38	<0.001
ME, kcal/kg	2485.6 [d]	2498.6 [c]	2535.3 [b]	2562.2 [a]	2561.4 [a]	2561.6 [a]	7.75	<0.001

WCON, 10 g pure water; SY10-1, 10 g sour yogurt + 10 g distilled water; SY20-1, 20 g sour yogurt + 20 g distilled water; SY30-1, 30 g sour yogurt + 30 g distilled water; SY40-1, 40 g sour yogurt + 40 g distilled water; SY50-1, 50 g sour yogurt + 50 g distilled water; ELOS, solubility of the organic matter in cellulase; EULOS, insoluble organic matter in cellulase;DOM,digestible organic matter; SEM, standard error of the mean. [a–e] Means with different letters in the same line are statistically significant ($p < 0.05$).

According to the results of the Pearson's correlation analysis (Table 13), in Trial I, a strong correlation was found between the pH and *Lactobacilli* counts (r= −0.852, $p < 0.001$), DM loss (r = −0.662, $p < 0.001$), OM (r = 0.740, $p < 0.001$) and ADF (r = −0.842, $p < 0.001$) on the day the silages were opened. In Trial II, the strong correlation between the *Lactococci* and opening day yeast (r = −0.724, $p < 0.001$) and NH_3-N (r = 0.572, $p < 0.05$) was remarkable. Significant correlations were found between the DM loss and DOM (r = −0.605, $p < 0.01$) and ME (r = −0.553, $p < 0.05$), CP and DOM (r = 0.665, $p < 0.01$) and ME (r = 0.722, $p < 0.001$), and ADF and DOM (r = −0.584, $p < 0.05$) and ME (r = −0.563, $p < 0.05$) (Table 13).

Table 13. Pearson's correlations of the variablefor experiments I, II.

Variable	1	2	3	4	5	6	7	8	9
Experiment I									
Lactobacilli	−0.852 **	0.948 **	−0.842 **	0.488 *	−0.862 **	0.426	−0.942 **	0.231	−0.077
Lactococci	−0.252	−0.029	−0.261	0.635 **	−0.106	−0.889 **	−0.029	−0.101	−0.266
Dry matter	−0.008	−0.115	0.233	0.616 **	0.095	−0.711 **	0.327	−0.132	−0.124
DM loss	−0.662 **	−0.640 **	−0.662 **	0.447	−0.734 **	0.011	−0.832 **	0.023	−0.212
OM	0.740 **	−0.762 **	0.699 **	−0.549 *	0.730 **	−0.110	0.741 **	−0.418	0.006
CP	−0.304	0.552 *	−0.286	−0.124	−0.529 *	0.734 **	−0.618 **	0.295	0.172
NDF	−0.255	0.386	−0.178	−0.140	−0.449	0.693 **	−0.625 **	−0.106	−0.254
ADF	−0.842 **	0.826 **	−0.785 **	0.583 *	−0.842 **	0.032	−0.893 **	0.307	−0.059
Experiment II									
Lactobacilli	−0.206	−0.370	−0.781 **	0.634 **	−0.373	−0.829 **	0.042	0.457	0.411
Lactococci	0.110	−0.422	−0.698 **	0.706 **	−0.724 **	−0.719 **	0.572 *	0.371	0.261
Dry matter	0.009	0.403	−0.226	0.091	0.164	−0.32	0.159	0.391	0.450
DM loss	−0.134	−0.251	0.462	−0.628 **	0.498 *	0.057	0.008	−0.605 **	−0.553 *
OM	−0.246	0.548 *	0.487 *	−0.543 *	0.792 **	0.587 *	−0.483 *	−0.128	0.034
CP	−0.067	0.454	−0.449	0.282	0.036	−0.125	−0.398	0.665 **	0.722 **
NDF	−0.064	0.159	0.386	−0.530 *	0.628 **	0.218	0.309	−0.300	−0.216
ADF	−0.105	−0.477 *	0.263	−0.434	0.273	−0.236	0.137	−0.584 *	−0.563 *

DM loss, dry matter loss; OM, organic matter; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber; 1, pH; 2, after aerobic exposure pH; 3, WSC; 4, LA; 5, yeast; 6, after aerobic exposure yeast; 7, NH_3-N; 8, DOM; 9, ME; * $p < 0.05$, ** $p < 0.01$.

4. Discussion

According to the physical evaluation, the addition of sour yogurt resulted in forming green silages with a very pleasant odor and intact stem and leaf integrity. Compared to the study of Kiani et al. [16], it was seen that our findings were higher, while the CON and WCON groups scored close to Kiani et al. [16]. It was found that the Flieg scores of all the groups were higher than those of Kiani et al. [16], while the SY30-2 group had the highest Flieg score. The addition of sour yogurt had a positive effect on the odor of the silages, especially the odor of the SY20-1, SY20-2, SY30-1, and SY30-2 groups, suggesting that the palatability was very high. This was due to the homofermentative LABs (*L. bulgaricus* and *S. thermophilus*) in the yogurt increasing LA production. The high amount of LA in the yogurt groups supported this.

Kiani et al. reported that adding 5% sour yogurt to corn silages caused an increase in the amount of CP compared to the CON group and a decrease in NH_3-N [16]. Except for SY10-1, the amount of CP increased similarly to Kiani et al. in all yogurt added groups [16]. Despite the increased sour yogurt in corn silages, NH_3-N decreased, which was similar to that of Kiani et al. [16]. However, Meeske et al. [26] stated that adding lactic acid bacterial inoculant (*Lactobacillus plantarum*, *Pediococcus acidilactici*) decreased the CP, caused the NH_3-N to below in the inoculant group and not supported the decrease in CP. Sour yogurt can be an alternative to commercial lactic acid bacteria inoculants because it increases the CP of corn silages and decreases NH_3-N, slowing down proteolysis, despite increasing the ratio. In addition, *S. thermophilus* has limited proteolytic activity and requires free amino acids for growth. These are glutamic acid, histidine, cysteine, methionine, valine, leucine, isoleucine, tryptophan, arginine and tyrosine. However, the free amino acids naturally found in milk are not enough. Free amino acids are supplemented using the short-chain peptides produced during heat treatment in milk or by the breakdown of milk proteins by *L. bulgaricus* [35]. The amino acids and peptides that emerged during proteolysis in the silages were used by *S. thermophilus*, and therefore, the amount of NH_3-N might have been found to be low, especially in groups to which yogurt was added at high rates. This will increase the low CP amount of corn silages and contribute to meeting the CP requirement of ruminants.

The addition of different ratios of sour yogurt caused an increase in the CA contents of corn silages relative to the CON group in Trial I. This increase was due to the 0.97% CA (natural) content of yogurt. However, in Trial II, the amount of CA in the yogurt groups was found to be close to the WCON group, and it was found to increase only in the SY10-1 group. However, Kiani et al. determined a decrease in CA, which is inconsistent with our results [16]. In Trial II, there was an increase in DM loss since the amount of water was higher in the groups to which sour yogurt was added. This explains why CA did not increase, despite the increase in the amount of yogurt.

In Trial I, the NDF was similar to the CON in the SY10-2 and SY30-2 groups, while the NDF and ADF of other yogurt groups increased compared to the CON. Kara et al. reported that the addition of homofermentative LAB increased the NDF and ADF contents of corn silages [36]. In Trial II, the NDF of SY10-1 was similar to the WCON; the NDFs of SY20-1, SY30-1, and SY40-1 were low, and the ADFs of SY30-1 and SY50-1 were low. Especially in the SY50-2 and SY50-1 groups, the increase in the NDF contents is consistent with that of Kara et al. [36]. This may be due to the high DM losses in the SY50-2 and SY50-1 groups. Marbun et al. [37] reported that different lactic acid bacteria inoculants did not significantly affect the NDF and ADF in corn silages. Sour yogurt, which we consider an alternative to bacterial inoculants, did not show any effect on NDF and ADF, in agreement with Marbun et al. [37] and other studies [38–40]. It has been reported that DM varies between 16.87–23.56% in the corn varieties grown as a second crop, and DM is 20.37% in the Pioneer-3167 cultivar [41]. Sabia et al. reported 7.23% CP and 6.55% CA in DM in the whole-plant corn of the Pioneer-PR32W86 cultivar [42]. In the research, Pioneer 32K61 corn, grown as the second crop, containing 23.70% DM, 7.8% CP of DM and 6.2% CA of DM, was used as the starting material. In this study, sour yogurt added to corn at increasing rates and different dilution levels by significant increased the DM contents in the SY30-2 and SY30-1 groups. Kiani et al. reported that sour yogurt does not affect DM. A rapid decrease in the pH level is required to obtain high-quality, well-fermented palatable silages and inhibit the growth of enterobacter and clostridia [16].This is made possible by producing lactic acid by homofermentative lactic acid bacteria using water-soluble carbohydrates [26]. In Trials I and II, the pHs of all groups were found to be between 3.51 and 3.63, which were numerically lower than Kiani et al.'s [16], and in the range of 3.5–4.0 pH [43], which were reported for good quality silages. In addition, a low pH in silages is related to the success of the silage during silage processing and storage [19]. In Trials I and II, the WSC decreased, and LA increased with the increasing sour yogurt levels. Polat et al. reported that the amount of LA in the corn silages silaged with additives containing *Lactobacillus* *plantarum* and *Enterococcus faecium* was 2.21% [44]. In general, lactic acid is the end product of fermentation in silage due to its strong acid (pKa 4.76) properties [19]. In our study, high lactic acid concentrations caused a rapid decrease in the pH. This reduced the activity of harmful microorganisms and the production of butyric acid.

When the yogurt used in the research was placed on the market shelf, the WSC content was determined as 114.08 g/kg DM and the LA as 157.87 g/kg DM. Souring takes place in about 3–5 days. Since consumers do not prefer the sour taste in yogurt, even if the shelf life is not completed, it is separated to be thrown away. The WSC content of sour yogurt was determined as 90.29 g/kg DM, and the LA amount was determined as 115.95 g/kg DM. In traditional and industrial yogurt production, two lactic acid bacteria species, *L. bulgaricus* and *S. thermophilus*, are widely used [45]. Yogurt bacteria, *L. bulgaricus* [46] and *S. thermophilus* [46,47] are known as homofermentative LAB. As in homofermentative inoculants [7,8], the addition of sour yogurt successfully shifted the fermentation to LA, AA ratios decreased due to increasing yogurt levels, and an increase in the LA/AA ratio occurred, which is expected. The increase in ethanol with the increase in (Trial I, II) sour yogurt level may be due to the high ethanol content in sour yogurt.

Compared to the CON and WCON groups, the NH_3-N content of the corn silages decreased similarly to that of Kiani et al., despite the increase in the amount of sour yogurt [16]. Differently diluted sour yogurt was effective in inhibiting proteolysis. This effect is particularly evident in the SY30-2, SY40-2, SY20-1, and SY30-1 groups. Sour yogurt decreased the pH of silage and inhibited the multiplication of microorganisms, which caused proteolysis and degraded native plant proteins to non-protein N. While sour yogurt effectively prevented DM loss in the SY30-2 and SY30-1 groups, it could not prevent DM loss in other groups, similar to that of Kiani et al. [16].

Meeske et al. reported that the 90th-day *Lactobacilli* counts were not affected; however, the *Lactococci* counts increased in the corn silages using LAB inoculants [26]. It was found that sour yogurt increased the *Lactobacilli* counts in Trials I and II, unlike Meeske et al. [26], while it increased the *Lactococci* counts in SY10-2, SY20-2, and Trial II, similar to that of Meeske et al. [26]. In the PY40-2 and PY50-2 groups, a decrease was determined in the *Lactobacilli* counts compared to the CON group. The yeast counts were reduced by ensiling relative to the starting material, but the low pH was unable to inhibit yeast growth, similar to that of Meeske et al. [26]. In addition, the findings of this study are consistent with the yeast counts determined by Kara et al. in the maize silages opened on the 45th day of homofermentative LAB addition [36]. Ensiling prevented the growth of enterobacteria in the SY20-2 group. Mold could not be determined in the groups; in addition, Caicedo and Caicedo reported that harmful microorganisms did not grow during the 30 days of fermentation in cocoa shell silage and mango silage treated with natural yogurt [48,49]. When the aerobic stability results were evaluated, we determined that the SY10-2, SY20-2, SY10-1, and SY20-1 groups were more durable than the others. In the aerobic period, there was a very rapid growth of yeast, and the yeast counts of all groups were found to be above the hygienic risk limit (5 CFU/g). The homofermentative properties of yogurt bacteria increased the LA content in silages and decreased aerobic stability. This is an expected situation, as in homofermentative LAB inoculants.

Meeske et al. reported that the inoculants increased the in vitro DOM content of corn silages numerically [26]. However, studies also report that LAB inoculants do not affect dry matter digestibility (DMD) [37,50]. Weinberg et al. showed that LAB inoculants could potentially ameliorate DMD, improving animal performance [51]. Reyes-Gutiérrez reported that sugarcane silage with a bacterial inoculum containing 3% and 1% additives has a higher in vitro organic matter digestibility [52]. In this study, ELOS, DOM, and ME increased in the SY30-2, SY40-2, and SY50-2 groups in Trial I and all yogurt groups in Trial II. Sour yogurt improved the DOM and ME contents of corn silages, similar to those of Weinberg et al. [52].This may be due to the increased amount of protein in the silage because yogurt (natural) contains CP at 5.48% DM. Increasing yogurt caused an increase in ELOS and a decrease in EULOS. Depending on these, DOM and ME improved.

5. Conclusions

The addition of different levels of diluted sour yogurt improved the fermentation quality of second-crop corn silage and increased the LAB counts and efficiency by promoting LAB development. Additionally, proteolysis was inhibited, the degradation of proteins to ammonia was reduced, and the digestibility of organic matter and ME contents increased. Hence, sour yogurt (20 and 30 g/kg fresh corn levels) could be used in the preparation of second-crop corn silage. However, the effect of sour yogurt on aerobic stability was poor, as with homofermentative LAB inoculants. The utilization of sour yogurt is effective and economical for improving the fermentation quality, DOM and ME of second-crop maize silage and reducing protein degradation. Further studies are advised to use different plants to reveal the mechanisms during ensiling.

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