Evaluation of microbial and sensory properties of flavored yogurt drink produced by *Noanea mucronata* and liquid smoke treatment

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Abstract

Liquid smoke is more acceptable compared with traditional smoking for various practical and health reasons. This study aimed to investigate the quality attributes of yogurt drink treated with natural and liquid smoke. Yogurt drink samples were divided into four groups; natural smoked, liquid smoked at two concentrations (1 and 2 $mL \cdot L^{-1}$) and un-smoked control samples. Microbial and sensory attributes of yogurt drink samples were analyzed after 1, 7, 14, and 21 days of storage at 4°C.

The final counts of total bacteria, lactic acid bacteria, yeast and molds were significantly lower after the application of 2 $mL \cdot L^{-1}$ liquid smoke compared to control samples. Moreover, lactic acid bacteria of yogurt drink were not inhibited by natural smoke or 1 $mL \cdot L^{-1}$ liquid smoke treatments. According to the sensory evaluation and microbiological analysis, application of 1 $mL \cdot L^{-1}$ of liquid smoke in yogurt drink is the most appropriate and convenient alternative to the traditional smoking method.

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Introduction

There is a growing demand for yogurt drinks due to their unique properties and numerous health benefits.[1] Yogurt drinks are rich in potassium, calcium, protein and B vitamins, which help stabilize the immune system.[2] In addition, yogurt drinks are beneficially ameliorated the behaviour and well-being of the consumers.[3,4] Yogurt drink is known by various names in many regions such as Ayran in Turkey, Dahi and Lassi in India, Laban in Arab countries, and Doogh in Iran.[5]

Yogurt drink beverage is popular in Armenia, Iran, Azerbaijan, Turkey, Afghanistan, Iraq, Syria, and Balkans.[6] Smoked yogurt drink is a popular kind of flavored yogurt drink that is manufactured by traditional methods in Kurdistan, Iran (Middle East, Western Asia). Its production is based on mixing prepared yogurt drink with smoke from thorny saltwort (*Noaea mucronata*).[7]

Noaea mucronata belongs to the family of Chenopodiaceae which is spread on arid and semi-arid rangelands of Middle East, North Africa and some part of Europe. The plant shrub has the sprawling branched stems terminating in sharp spines and leaves are narrowly-linear, mucronate, and slightly decurrent at the base.[8] *N. mucronata* is commonly abounded in Mediterranean climates, with long hot dry summers and cold winters. It appears that the plant can survive a climate that's not Mediterranean at the eastern border of its native range in Iran.[9] The dry matter of *N. mucronata* was found to contain 40.0 % nitrogen-free extract, 23.0 % dietary fiber, 13.5 % total protein, 2.1 % ash, and 2.3 % total fat.[10]

Farag *et al.*[11] showed in a study that *N. mucronata* contained 16 amino acids with the highest and lowest concentration of free amino acids being aspartic acid and histidine, respectively. They also reported that Arachidic acid is the major fatty acid of *N. mucronata*.

Also, Pharmacological effects of *N. mucronata* have been widely studied in Egypt, Saudi Arabia and Iran.[12-14] *N. mucronata* has been used as a treatment of kidney stones in Iran.[12] The plant is also widely used as a fuel for cooking and heating.[10]

Liquid Smoke (LS) is often preferred over traditional smoking

because of omission of carcinogens, ease of use, greater uniformity, and repeatability of the production.[15] Consequently, it can be applying as a safe alternative to food protective without mutagenic[16] and carcinogenic materials.[17] Liquid smoke is considered a natural flavouring and has been granted Generally Recognized as Safe (GRAS) status by the Flavor and Extract Manufacturers Association.[18]

Smoke applications in addition to imparting flavor, color and aroma to foods, have been used for food preservation because of their antimicrobial and antioxidant properties.[19] Several studies were conducted on antibacterial activity of smoke applications in food products.[19-24] Gonulalan *et al.*[22] compared the effect of liquid smoke and traditional smoking on beef tongue. Gedela *et al.*[21] studied the effect of liquid smoke on reduction of *Listeria monocytogenes* in frankfurters. Other researchers introduced LS flavouring as an alternative to traditional smoking of fish fillets; also, they found that the sensory properties of liquid smoked fish were as good as traditional smoked fillets.[23]

Even though many studies have reported the use of Natural Smoke (NS) and LS in food, there has been no published research on the effectiveness of smoke treatments in yogurt drinks. The objective of the present study is to ascertain effect of LS in comparison with NS on microbial and sensory attributes of yogurt drink during storage.

Materials and Methods

Smoke and yogurt materials

Hickory liquid smoke was obtained from the Reily Foods Company (New Orleans, LA, USA). According to the information published by the manufacturer the Hickory Liquid Smoke is approved by the European Parliament (EC) No 1321/2013 and 2065/2003 as a smoke flavoring agent. Traditional yogurt and dried *N. mucronata* plants were purchased from a local dairy store in Sanandaj, Kurdistan, Iran (35°18′52″N 46°59′32″E).

Production of yogurt drink

Yogurt drink samples were prepared by mixing the traditional yogurt (fat=1.65 %, dry matters=8.63 ± 0.1%) and distilled water at a ratio of 50:50 v/v using a mixer (Type SM, Sanyo Electric Co., Ltd, Japan) at 25 °C, then 0.01 w/v salt (NaCl; Merck, Darmstadt, Germany) was added to this mixture.

Production of smoke-flavored yogurt drink

After preliminary sensory experiments, the appropriate concentrations of liquid smoke (1 and 2 mL·L^{-1}) were determined. Therefore, yogurt drink samples were divided into four groups; 1: unsmoked or control; 2: traditionally smoked yogurt drink; 3: 1 mL·L^{-1} liquid smoke added; and 4: 2 mL·L^{-1} liquid smoke added

For the traditional smoking the protocol for preparing traditional smoked yogurt drink samples was provided by a local dairy store. 150 g of *N. mucronata* plant was burnt for 10 s and the flames were extinguished by covering with an aluminum foil. The remaining plant material was placed in an empty plastic container carefully. The container was allowed to fill with the smoke for 2 min. Then the plant material was removed, and 5 liters of yogurt drink sample was added to make the traditional smoked samples.

Yogurt drink samples were stored for 24 h at 4±1ºC to allow equilibration. Microbial and sensory attributes of yogurt drink samples were examined after 1, 7, 14, and 21 days of storage at 4°C.[24] The experiments were conducted for three independent batches and all microbial and sensory tests were performed in triplicates.

Measurement of TA and pH

The pH and Titratable Acidity (TA) of the resulting yogurt drink samples were measured using an Accumet® Research AR15 pH meter (Fisher Scientific, Kalamazoo, Michigan, USA), and the titration was done using NaOH (0.1 mol·L^{-1}) in the presence of phenolphthalein as an indicator.[25]

Microbiological analysis of yogurt drink samples

First, ten milliliters of each sample was pipetted aseptically into 90 mL of quarter strength Ringer's solution and mixed thoroughly. Serial dilutions of samples (10^{-1} to 10^{-8}) were prepared. After serial dilutions, Total Counts of Bacteria (TCB) were determined on Plate Count Agar (PCA; Merck, Darmstadt, Germany), after incubation at 30°C for 72 h. Coliforms were enumerated on Violet Red Bile Agar (VRBA; Merck), after incubation at 32ºC for 72 h. The yeasts and molds were counted on Yeast Extract Glucose Chloramphenicol Agar (YGC; Merck) after five days of incubation at 25ºC.[25] Total counts of Lactic Acid Bacteria (LAB) were enumerated on de Man, Rogosa and Sharpe agar (MRS; Merck), the plates were incubated in anaerobic jars under carbon dioxide-nitrogen gas atmosphere (GasPak System; BBL, Cockeysville, MD, USA) at 30 °C for 72 h.[25] The enumeration of *Staphylococcus aureus* and *Escherichia coli* was done using the surface spread method on Baird-Parker agar (BP; Oxoid, Basingstoke, England) and Sorbitol–MacConkey agar (SMAC; Oxoid), respectively.[25] The plates for *E. coli* were incubated at 44 °C and for *S. aureus* were incubated at 37 °C for 72 h.[25] Besides, biochemical confirmation tests for detecting *E. coli* and *S. aureus* were completed.[25]

Sensory evaluation of smoked yogurt drink

Yogurt drink samples were evaluated for odor, taste, appearance (color and existence of dark particles) and overall acceptance. Fifty panelists 25 females and 25 males, ages between 20 and 48 were recruited among students and staff. Two digits coded samples at a temperature of 4°C with a volume of 50 mL inside the clear disposable plastic cups were presented to participants after 1, 7, 14, and 21 days of storage. Participants were asked to indicate how much they liked or disliked each product on a 5-point hedonic scale (5=like extremely, 4= Like moderately, 3= Neither like nor dislike, 2= Dislike moderately, and 1=dislike extremely). Mineral water (Aquafina, Tehran, Iran) was served for rinsing between tests. An evaluation was conducted under bright lighting and at 25°C of room temperature.[26]

Statistical analysis

The obtained data were subjected to statistical analysis using a one-way ANOVA using SAS software version 9.1 (SAS Institute, Cary, NC, USA). The General Linear Model Repeated Measures was used to determine the differences between each period, and the differences between means for each parameter were determined using Duncan's multiple range test. Differences were accepted when $P<0.05$.

Results

TA and pH evaluation of yogurt drink samples

According to Table 1 the effects of smoke treatments on the TA

and pH of yogurt drink samples were not significantly different (P>0.05). However, in all treatments TA increased significantly during storage time (P<0.05) as the pH of the samples decreased significantly from ~4.3 to 4.0 at the end of storage.

Microbiological evaluation of yogurt drink samples

As shown in Table 2, smoke treatments did not have any significant effects (P>0.05) on LAB counts, coliform, yeast and mold and *Staphylococcus spp*. after one day of yogurt drink preparation. The coliform counts were not observed in any yogurt drink samples after seven days of storage. Furthermore, *E. coli* and *S. aureus* were not detectable in any yogurt drink samples.

LAB counts in yogurt drink samples treated with 2 mL·L^{-1} liquid smoke decreased significantly from 7.01 to 6.79 log CFU·mL^{-1} during 14 days and were followed by a significant drop to the 6.28 log CFU·mL^{-1} at the end of storage time. Although

Table 1. TA and pH change of yogurt drink samples during storage time.

	1d	*7d*	*14d*	*21d*
TA				
Control	0.603±0.013a, D	0.621±0.011a, C	0.648±0.013a, B	0.666±0.026a, A
N.S	0.603±0.027a, D	0.621±0.025a, C	0.648±0.035a, B	0.666±0.025a, A
1 mL·L^{-1} LS	0.603±0.018a, B	0.617±0.016a, B	0.644±0.032a, A	0.657±0.036a, A
2 mL·L^{-1} LS	0.603±0.014a, B	0.612±0.013a, B	0.639±0.024a, A	0.648±0.045a, A
pH				
Control	4.312±0.013a, A	4.244±0.012a, B	4.17±0.011a, C	4.064±0.012a, D
N.S	4.313±0.017a, A	4.241±0.056a, B	4.16±0.035a, C	4.073±0.023a, D
1 mL·L^{-1} LS	4.324±0.012a, A	4.242±0.034a, B	4.17±0.033a, C	4.063±0.035a, D
2 mL·L^{-1} LS	4.314±0.036a, A	4.243±0.047a, B	4.17±0.031a, C	4.064±0.037a, D

Values are means ±SE and values followed by the same capital letters in each row (Effect of storage time) or the same small letters in a group of a column (effect of different smoke treatments), are not significantly different at P<0.05. d: days of storage. All tests were performed in triplicate for three independent experiments.

Table 2. Microbial changes (log CFU·mL^{-1}) of yogurt drink samples during storage time.

	1d	*7d*	*14d*	*21d*
TCB				
Control	7.86±0.038a, A	7.85±0.014a, A	7.63±0.042a, B	7.43±0.014a, C
N.S	7.86±0.034a, A	7.79±0.028a, B	7.21±0.014b, C	6.90±0.023b, D
1 mL·L^{-1} LS	7.86±0.024a, A	7.75±0.024a, B	7.21±0.056b, C	6.67±0.046c, D
2 mL·L^{-1} LS	7.88±0.032a, A	7.51±0.037b, AB	6.99±0.024c, B	6.30±0.055d, C
LAB				
Control	7.04±0.065a, A	7.02±0.047a, A	7.01±0.025a, A	6.92±0.014a, B
N.S	7.04±0.066a, A	6.99±0.048a, AB	6.95±0.002b, B	6.83±0.013b, C
1 mL·L^{-1} LS	7.04±0.087a, A	6.98±0.004a, AB	6.93±0.024b, BC	6.84±0.001b, C
2 mL·L^{-1} LS	7.01±0.034a, A	6.91±0.003b, B	6.79±0.003c, C	6.28±0.031c, D
Staphylococcus spp.				
Control	1.87±0.03a, A	1.85±0.046a, A	1.63±0.056a, B	1.20±0.031a, C
N.S	1.86±0.03a, A	1.79±0.043ab, B	1.38±0.043b, C	<1.0×101b, D
1 mL·L^{-1} LS	1.86±0.02a, A	1.75±0.045b, B	1.24±0.057c, C	<1.0×101b, D
2 mL·L^{-1} LS	1.88±0.03a, A	1.29±0.034c, A	<1.0×101d, B	-
S. aureus				
Control	-	-	-	-
N.S	-	-	-	-
1 mL·L^{-1} LS	-	-	-	-
2 mL·L^{-1} LS	-	-	-	-
Coliform				
Control	1.36±1.085a	-	-	-
N.S	1.38±0.074a	-	-	-
1 mL·L^{-1} LS	1.39±0.044a	-	-	-
2 mL·L^{-1} LS	1.37±0.043a	-	-	-
E. coli				
Control	-	-	-	-
N.S	-	-	-	-
1 mL·L^{-1} LS	-	-	-	-
2 mL·L^{-1} LS	-	-	-	-
Yeast and mould				
Control	1.80±0.013a, C	1.87±0.012a, B	1.90±0.002a, AB	1.99±0.013a, A
N.S	1.91±0.018a, A	1.90±0.025a, A	1.89±0.024a, A	1.90±0.025b, A
1 mL·L^{-1} LS	1.81±0.016a, A	1.79±0.027b, A	1.76±0.026b, AB	1.76±0.013c, B
2 mL·L^{-1} LS	1.80±0.014a, A	1.74±0.023c, AB	1.66±0.023c, BC	1.61±0.014d, C

Values are means ±SE and values followed by the same capital letters in each row (effect of storage time) or the same small letters in a group of a column (effect of different smoke treatments), are not significantly different at P<0.05. d: days of storage. All tests were performed in triplicate for three independent experiments.

LAB counts in control samples remained stable at 7 log CFU·mL^{-1}, a slight decrease was observed in LAB populations in samples treated with traditional smoke and 1 mL·L^{-1} liquid smoke during 14 days of storage (P<0.05). After 21 days of storage, LAB counts significantly decreased to 6.84, 6.83 and 6.92 log CFU·mL^{-1} in yogurt drink treated with 1 mL·L^{-1} liquid smoke, NS and control samples, respectively (P<0.05).

The populations of *Staphylococcus spp.* in all samples decreased significantly (P<0.05) during storage time in all samples. After seven days of storage, the count of *Staphylococcus spp.* in 2 mL·L^{-1} liquid smoked yogurt drink, 1 mL·L^{-1} liquid smoked yogurt drink and natural smoked samples were 0.56, 0.1 and 0.06 log CFU·mL^{-1} lower than the control sample. During 14 days of storage, the *Staphylococcus spp.* counts decreased significantly to 1.63, 1.38, 1.24 and <1 log CFU·mL^{-1} in control, natural smoked yogurt drink, 1 mL·L^{-1} and 2 mL·L^{-1} liquid smoked yogurt drink, respectively. In the following seven days, the *Staphylococcus spp.* counts decreased significantly to 1.20 in control, and less than 1 log CFU·mL^{-1} for all other samples respectively. Yeast and mold counts in control samples increased significantly from 1.80 to 1.99 log CFU·mL^{-1} during storage time; In contrast, in samples treated with 1 mL·L^{-1} and 2 mL·L^{-1} liquid smoke the counts of yeast and mold decreased from 1.81 to 1.76 and from 1.80 to 1.61 log CFU·mL^{-1}, respectively. Yeast and mold populations in samples treated with traditional smoke remained stable during storage. The TCB of all yogurt drink samples decreased significantly (P<0.05) during storage. After seven days of storage, the TCB of 2 mL·L^{-1} liquid smoked yogurt drink, 1 mL·L^{-1} liquid smoked yogurt drink and natural smoked yogurt drink were 0.34, 0.1 and 0.06 log CFU·mL^{-1} lower than control samples. After 14 days, no significant differences were observed in TCB of 1 mL·L^{-1} liquid smoke and traditional smoked yogurt drink samples. On the other hand, the TCB in 2 mL·L^{-1} liquid smoke was 0.64 log CFU·mL^{-1} lower than TCB of control samples. At the end of 21 days, 2 mL·L^{-1} liquid smoke with 6.30 log CFU·mL^{-1} had a lower number of TCB.

Sensory evaluation of yogurt drink samples

Table 3 presents the sensory attributes of yogurt drink samples during storage. A day after preparation, there were no significant differences regarding taste among samples (P>0.05). However, the samples treated with smoke had significantly higher odor scores. The natural smoked yogurt drink samples had a significantly lower score regarding appearance, and this downward trend was observed until the end of storage time. After seven days, the unsmoked control samples had significantly lower mean taste and odor score (P<0.05). No statistical differences were observed (P>0.05) between taste and odor scores of traditional smoked samples and liquid smoke samples after 7 days. After 14 days, the taste and overall acceptance of 2 mL·L^{-1} smoked yogurt drink samples and control samples had significantly higher and lower mean scores, respectively (P<0.05). At the end of 21 days, control samples had significantly lower taste and overall acceptance scores. However, there were no significant differences between traditional smoked samples and 1 mL·L^{-1} liquid smoked yogurt drink samples score of overall acceptance (P>0.05). In general, the sensory attributed of all samples were dropped significantly during 21 days of storage. However, samples treated with 2 mL·L^{-1} liquid smoke had significantly higher taste and odor scores compared with other samples.

Discussion

There has been very little research on microbiology and sensory of smoked dairy products.[20] To the best of our knowledge this is the first study investigated effects of regular and liquid smoke treatments on microbial and sensory of yogurt drink samples. Thus, comparisons will be made with the results of similar studies. It is well known that metabolism of carbohydrates and formation of organic acids in yogurt contribute to the sour taste and result in pH decrease.[27] The pH values for Iranian yogurt drink should be less than 4.5.[6] Similarly, various pH values ranging from 3.44 to 4.44 have been reported for Ayran (Turkish fermented

Table 3. Sensory attribute changes of yogurt drink samples during storage time.

	1d	7d	14d	21d
Taste				
Control	4.60±0.4949a,A	4.08±0.3405b,B	3.18±0.3881c,C	2.22±0.4647c,D
N.S	4.64±0.4849a,A	4.40±0.4949a,B	3.60±0.4949b,C	2.80±0.4041b,D
1 mL·L^{-1} LS	4.62±0.4903a,A	4.44±0.5014a,A	3.58±0.4986b,B	2.90±0.5440ab,C
2 mL·L^{-1} LS	4.50±0.5051a,A	4.42±0.4986a,A	4.04±0.4932a,B	3.08±0.5657a, C
Odor				
Control	4.10±0.5051b,A	4.00±0.4518b,AB	3.66±0.4785c,B	2.52±0.5436b, C
N.S	4.46±0.5035a,A	4.38±0.4903a, A	3.94±0.4699b,B	2.88±0.4352b, C
1 mL·L^{-1} LS	4.38±0.4903a,A	4.32±0.4712a,AB	4.16±0.4219a,B	3.36±0.4849a, C
2 mL·L^{-1} LS	4.28±0.4536ab,A	4.24±0.4314a,AB	4.10±0.3642ab,B	3.40±0.4949a, C
Appearance (colour + existence of soots)				
Control	4.74±0.4431a, A	4.58±0.4986a, A	4.58±0.4986a, A	4.30±0.4629a,B
N.S	3.76±0.5175b, A	3.42±0.5379 b, B	3.38±0.5303b, B	3.28±0.4965b, C
1 mL·L^{-1} LS	4.68±0.4712a, A	4.54±0.5035a, A	4.50±0.5440a, A	4.24±0.5175a, B
2 mL·L^{-1} LS	4.60±0.4949a, A	4.48±0.5047a,AB	4.44±0.5014a, B	4.24±0.4314a, B
Overall acceptance				
Control	4.26±0.4431b, A	4.08±0.2740c, B	3.28±0.4536c, C	2.18±0.4819c, D
N.S	4.32±0.5127ab,A	4.22±0.4647bc,A	3.66±0.4785b, B	2.76±0.4314b, C
1 mL·L^{-1} LS	4.50±0.5440a, A	4.44±0.5406a, A	3.60±0.4949b, B	2.86±0.5349ab,C
2 mL·L^{-1} LS	4.44±0.5014ab,A	4.48±0.4903ab,A	4.08±0.4445a, B	3.04±0.5700a, C

Values are means ±SE and values followed by the same capital letters in each row (effect of storage time) or the same small letters in a group of a column (effect of different smoke treatments), are not significantly different at P<0.05. d: days of storage. All tests were performed in triplicate for three independent experiments.1 (Dislike extremely), 2 (Dislike moderately),3 (Neither like nor dislike), 4 (Like moderately),5 (Like extremely).

yogurt drink).[28]

Yeasts and molds are abundant in environments due to their ability to tolerate to low pH and temperature values and ability to utilize a variability of substrates.[29]

The results showed that the number of yeasts and molds slightly increased during storage. This might be due the acidic resistance of yeast that is reliant on the energy requiring system in cells that pump protons dynamically out of the cells and thus prohibit acidification of the cell interior; Therefore, yeasts are quite adaptive to incompatible conditions such as acidity.[30] Moreover, the slight increase in yeast counts of control yogurt drink samples can be attributed to the nutritional profile and low pH of yogurt drinks, which are favorable for the growth of spoilage.[31] According to the Iranian regulations for pasteurized yogurt drink the counts of coliform, yeast and molds should be less than 10 and 100 CFU·mL^{-1}, respectively, and the yogurt drink should be free of *E. coli* and *S. aureus*.[6] Mehraban *et al.*[32] reported up to 5.7×10^2 CFU·mL^{-1} of coliforms and 3×10^7 CFU·mL^{-1} for LAB in yogurt drink samples, these counts are similar to our findings of LAB and coliform counts; however, they reported higher yeast counts (1.2×10^5 CFU·mL^{-1}) in comparison to our results. Furthermore, the LAB counts at the end of storage was in agreement with reported results by Birollo *et al.*[33] which indicated counts up to 10^7 CFU·g^{-1} in yogurt at the end of 30-day storage time. For yogurt products the minimum acceptable levels of LAB counts are established as10^6 CFU·g^{-1} in Switzerland and Italy, 10^7 CFU·g^{-1} in Japan and 10^8 CFU·g^{-1} in Portugal.[34] The LAB counts for 1 mL·L^{-1} LS treatment was around 10^7 CFU·mL^{-1} at the end of 21 days of storage and was consistently higher compared to LAB counts for the 2 mL·L-1 LS samples.

The presence of *Staphylococcus spp.* may indicate the poor sanitary and handling during the production and distribution of locally produced yogurt.[35] As pH values for yogurt drink samples decreased, the conditions became unfavorable for *Staphylococcus* spp. Thus, toward the end of the storage, *Staphylococcus* spp. were undetectable in yogurt.[35]

The decrease in TCB, LAB and *Staphylococcus spp.* counts over time for control samples may be related to nutrition limitations, as well as some organisms entering the death phase.[36]

The reductions in LAB counts for smoked yogurt drink samples are in agreement with the other study which showed that the inhibitory effect of liquid smoke on LAB increased with an increase in smoke concentration.[37] It has been proposed that phenols (Cresol, Guaiacol, Syringol, and Pyrocatechol) are the main components in liquid smoke responsible for the antimicrobial activity.[38] In agreement with results of our study, researchers indicated that the Staphylococcus were considerably more sensitive to liquid smoke compared to LAB.[39] The low pH of yogurt drink samples is the main reason of Staphylococcus inhibition. Also, the minimal values of pH for growth are influenced by other environmental factors.[40] The inhibition of *S. aureus* has been shown by applying 1 mL·L^{-1} acetic acid.[40] Therefore, the inhibition of *Staphylococcus spp.* using 2 mL·L^{-1} liquid smoke may be attributed to the amount of acetic acid contained in the liquid smoke.[41] The inhibition may also be due to the production of anti-staphylococcal compounds by the LAB.[42] Acidic stress and pH can damage cell membrane structures and reduce the activity of enzymes. Therefore, it seems that acidic stress and low pH may be the main reasons for the inhibition of staphylococci.[43] Coliforms have been reported to grow over a pH range of 4.4-9.0.[36] The inhibition of coliform growth probably is due to the low pH of yogurt drink samples. Similar results were reported that *E. coli* O157:H7 could not survive in the yogurt.[44] In contrast, other researcher found that coliforms can survive at pH of 3.67 in unheated yogurt drink samples.[26] Furthermore, no significant change in coliform counts was observed on the first day of smoke treatments suggesting that there is a lag time for smoke to be active on microorganisms in complex food systems.[38] Results of sensory analysis of current study are in agreement with earlier research in which they showed that the taste of yogurt drink samples changed during storage, as they showed the microbial metabolites, lipase, and ambient temperature are the most important factors for off- flavor of yogurt drink samples during storage time.[45] Also, a significant drop in sensory characteristics of fermented milk during storage time has been reported.[46]

The amount of acetaldehyde in yogurt drink samples probably decreased due to the degradation of acetaldehyde at lower pHs and enhanced oxidization during storage.[47] Furthermore, it can be suggested that 2 mL·L^{-1} liquid smoke can dominate and mask the off- flavor of yogurt drink at the end of storage. It is reported that phenolic compounds contribute to the smoke flavor of liquid smokes besides their antibacterial and antioxidant properties.[38]

The odor of smoked yogurt drink is due to volatile compounds present in the smoke condensate. Previous studies revealed that the most fraction of odor-active compounds in liquid smoke were alkyl and carbonyl derivatives of syringol and guaiacol.[48] In agreement with our results for liquid smoke samples, it showed that the sensory quality of liquid smoke treated samples were as good as that of traditional smoking.[49]

In conclusions, both sensory and microbial evaluations showed that excessive amount of liquid smoke is not suitable in yogurt drink products, therefore, the lower amount of liquid smoke can be the most advisable alternative to natural smoking to produce a safe and similar tasting yogurt drink product.

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