The Effect of Probiotic on Antioxidant Activity of Phenolic Compounds in Functional Yogurt Fortified by Plum Juice

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Abstract

Dairy products are suitable for functional meals since they have a high nutritional value. Yogurt is remarkable with its nutritional value over the other fermented dairy products. Fermented milk products help to maintain overall gastrointestinal health. The current study aimed to investigate the fortification of yogurt made of cow's milk and 10% plum (*Prunus domestica*) juice and culturing it with *bifidobacterium* as a probiotic along with regular culture *Lactobacillus delbrueckii subsp. bulgaricus* and *Streptococcus salivarius subsp. thermophilus* bacteria. Physicochemical, rheological, biochemical and microbial count tests were determined during storage times of 1,7 and 14 days at 4°C. The results showed that, the yogurt fortified with *Bifidobacterium* and plum juice had higher acidity, total solids (T.S.), and lower pH, fat, protein and ash compared to the control yogurt. All of the chemical composition parameters of T.S., fat, protein and ash percent decreased gradually over time of storage. The addition of 10% plum juice caused the highest syneresis amount and lowest viscosity of all treatments, which varied from 16.3 ml on day 1 up to 22.2 ml on 14 days and from (149±14 Centipoise (cP) at day 1 to (70±9cP) at day 14 and affected the overall acceptability of the yogurt and overall acceptability of all of the yogurt was affected with the storage period. Ellagic acid was the predominate (28.37±0.01 µg/ml) phenolic compound in plum juice, while in control yogurt was gallic acid, chlorogenic acid in probiotic yogurt, querectin in yogurt fortified with plum juice and catechin in probiotic yogurt fortified with plum juice. However, the addition of plum juice to yogurt has enhanced its total phenolic, total flavonoid content and antioxidant activity. However; this reduced the body and texture properties of yogurt. Furthermore, the addition of plum juice into the yogurt with or without *Bifidobacterium* had a lower bacterial count than without it. **Conclusion**: The current study results have indicated that yogurt supplement with plum juice using *bifidobacterium* and starter culture has many health benefits especially its antioxidant activity and phenolic compounds, which have protective effects against different diseases, on the other hand, it improves yogurt quality and gives customers a variety of choices.

Key word: Yogurt, plum juice, phenolic compound, antioxidant activity, probiotic, sensory evaluation, viscosity and synersis.

Introduction

Phenolic and polyphenolic compounds have great attention because of their beneficial health impacts on humans. There are abundantly found naturally in fruits and vegetables as secondary metabolites. These polyphenols are contributed to flavor, color, test, odor, bitterness, astringency and oxidative stability in foods (**Phaniendra *et al.*, 2015; Abdelshafy *et al.*, 2021 and Bondam *et al.*, 2022).** Furthermore, polyphenolic compounds were proven to have many pharmacological applications against osteoporosis, cardiovascular diseases, diabetes, neurodegenerative diseases, anticancer and antioxidant effects due to their chemical structure and their ability to react with lipids radicals and capturing the free radicals in the cells turning these radicals to stable products. Resulting in, stopping oxidative stress and cells damages **(Liguori *et al.,* 2018; Zhou *et al.,* 2020; Abdelshafy *et al.,* 2021; Mannucci *et al.,* 2021 and Bondam *et al.,* 2022).**

Functional foods are industrially produced or natural foods that, when consistently ingested within a diverse diet at effective levels, have potentially favorable benefits on health beyond basic nutrition (**Granato *et al.,* 2020).** Today's functional additives use *Lactobacillus*, which is widely used in industrial food manufacturing. Fermentation variables including pH, temperature, media composition, and others have an impact on *Lactobacillus's* ability to proliferate (**Widyastuti *et al.,* 2021**). Many bacteria are considered probiotics, but *Lactobacillus* and *Bifidobacterium* are the most common types used in foods because they have a long history of safe use and have been designated as GRAS (generally recognized as safe); in addition, they are the most prevalent types of bacteria found in the intestines of humans (**Nasef *et al.,* 2020.).** By adding specific probiotic bacteria strains to food products, studies have been conducted to provide evidence regarding the health benefits of probiotics on gastrointestinal infections, antimicrobial activity, improvement in lactose metabolism, serum cholesterol reduction, immune system stimulation, anti-mutagenicity, anti-carcinogenic, and anti-diarrhea activity **(Joseph, 2018).**

The FAO/WHO definition of a probiotic as “live microorganisms which when administered in adequate amounts, confer a health benefit to the host” **(Hill *et al.,* 2014, and Vitheejongjaroen *et al.,* 2021).** As a result, many medical and scientific studies have found that probiotic supplementation improves the body's antioxidant and anti-inflammatory capabilities (**Wang *et al.,* 2017; Moludi *et al.,* 2019, and Vitheejongjaroen *et al.,* 2021).** These antioxidants in both fruits and vegetables have the ability to give electrons to these unpaired electrons of ROS resulting in reversing their effects and promoting good health (**Gurnani *et al.*, 2014; Mahmoud *et al.*, 2017; Qureshi *et al.*, 2017, and Jideani *et al.*, 2021).**

Plum has considered one of the most valuable fruits grown worldwide. This fruit is so valuable for human nutrition as a result of its high energy, dietary, nutritional, and health values **(Jaroszewska, 2011 and, Yagmur and Taskin, 2011).** Also, plum has beneficial effects on human health due to their high content of antioxidants, phenols and anthocyanins **(Akbar, 2019; Johnson *et al.*, 2021, and Ma *et al.*, 2022).** So, both probiotics and fruits have beneficial impacts on human health due to their antioxidant capability, which reduces oxidative stress **(Queen and Tollefsbol, 2010; Sharifi-Rad *et al.*, 2020, and Vitheejongjaroen *et al.*, 2021)**.

In recent years, yogurt is considered one of the huge demand dairy products around the world when fortified with probiotics and fruits. According to **Ahmad *et al.*, (2022)**, yogurt is a popular dairy product prepared from milk fermentation with *Streptococcus thermophilus* and *Lactobacillus bulgaricus* bacteria. The addition of probiotic culture to this previous standard bacterial culture in preparing yogurt turns into probiotic yogurt, which has more antioxidant capability than regular yogurt **(Bchir *et al.*, 2019; Nyanzi *et al.*, 2021, and Akan *et al.*, 2022).** So, this work aims to investigate the physiochemical, biochemical, microbial, and functional properties of probiotic yogurt containing *Lactobacillus delbrueckii*, *Streptococcus salivarius* and *Bifidobacterium infantis* fortified by plum (*Prunus domestica*) juice, and effect of probiotic on antioxidant activity of phenolic compounds in functional yogurt.

Materials and methods

Whole cow's milk (3.50% fat, 13.59% total solids (T.S.), 3.82% protein, 0.94% ash) was obtained from Gemmeza Animal Production Research Stations, Gharbia governorate, Egypt.

All the bacterial strains *Streptococcus thermophiles* (*S. thermophiles)* EMCC 11044, *Lactobacillus bulgaricus (L. bulgaricus*) EMCC 11102, and *Bifidobacterium infantis* ATCC 11551, were obtained from Cairo Microbiological Resources Center, Egypt. These strains were transferred successively two times in skimmed milk (12% total solid) and were activated at 42 °C for 24 h before the yogurt production.

All the chemicals were purchased from El-Gomhoriya Company for Trading Drugs, Chemicals and Medical Instruments, in Egypt.

Plant material and juice preparation

Plum fruits were washed, and then the juice was obtained according to the method described by **Chavan *et al.*, (2013**). The juice was pasteurized at 90 °C for 15 sec then quickly chilled in an ice bath to below 10 °C and kept in sterilized glass bottles in the refrigerator (4°C) till use.

Yogurt manufacturing

The yogurt manufacturing was done according to procedures indicated by the "**International Dairy Federation's (IDF) yogurt manufacture procedures, (1988)** ", which started with heating the milk at 95 °C for 5 min, and then it was cooled to 42 °C and divided to the following eight formulas (three replicates each). Considering, 10 % of pasteurized plum juice was added and stirred quickly after the immediate addition of the 2 % (v/v, *S. thermophilus* and *L. bulgaricus* in 1:1 ratio starter culture) with or without *Bifidobacterium infantis* of

24 h old culture. Samples were incubated in a plastic cup at 42 °C (100 ml), until analysis then samples were stored at 4 °C.

Experimental treatments:

1st Treatment (Control yogurt): Milk inoculates with 2 % of yogurt culture, which is *L. bulgaricus* and *S. thermophilus* culture (1:1v/v).

2nd Treatment (Probiotic yogurt): Milk inoculates with 2 % of yogurt culture, which are *L. bulgaricus, S. thermophilus* and *Bifidobacterium infantis* culture (1:1: 0.3v/v).

3rd Treatment (Yogurt fortified with 10% plum juice): Milk inoculates with 2 % of *L. bulgaricus* and *S. thermophilus* culture (1:1v/v), with the addition of 10% plum juice.

4th Treatment (Probiotic yogurt fortified with 10% plum juice): Milk inoculated with 2% of *L. Bulgaricus, S. thermophilus* and *Bifidobacterium infantis* culture (1:1:0.3v/v), with the addition of 10 % plum juice.

All yogurt samples were incubated at 37 °C for 4-6 h until the pH reached 4.3 or 0.8 % acidity. Finally, yogurt samples were cooled and stored at 4°C until analysis.

Physicochemical analysis of yogurt

The physicochemical analyses include the following: pH, acidity as total titratable acid (TTA), total solids, moisture, fat, protein and ash were analyzed at an interval of 1, 7 and 14 days according to The Association of Official Analytical Chemists **(A.O.A.C, 1995)**. The pH measurement for all samples was done using a digital pH meter (Model pH-Kent EIL 7020). The TTA was determined according to **A.O.A.C (2000)** using sodium hydroxide 0.1mol/L. Then, the TTA of the yogurt samples was expressed as the lactic acid percentage.

Rheological Properties

- **Syneresis**

Syneresis of the yogurt treatments was determined by the volume of drained whey and estimated as a percentage of that volume using the method of **Lawrence, (1959).** The experiments were carried out three times/each. Eighty grams of the yogurt were placed on filter paper (Whatman No.1) on a glass funnel, which was placed on a graduated cylinder at room temperature for 2h for the syneresis index measuring. The syneresis index was measured from the collected liquid volume.

- **Viscosity**

DV-E Viscometer with No. 4 spindle (model LVDVE 230, Brookfield, serial No. E5896) was used for the determination of yogurt samples viscosity, and the results were expressed as complementary porridges (CPs) according to **Aryana, (2003**).

Sensory evaluation

After being kept in the refrigerator for 1, 7, and 14 days, yogurt samples were evaluated according to **Kasimoğlu *et al*., (2004),** by ten staff members (panelists) of Gemmeza Animal Production Research Stations, Gharbia governorate, Egypt. All samples were marked with codes and the order of presentation of samples was randomized for each panelist. Water was provided between samples as a palette cleanser and control yogurt served as the reference standard. The panelists rated body and texture, acidity, appearance, flavor and overall acceptability using a 100 scale.

Chemical analysis

-Determination of antioxidant activity

The antioxidant capacity of both fruits' juices and yogurt samples using the stable 2, 2-diphenyl-1-picrylhydrazyl radical

(DPPH•) radical, samples' capacity to scavenge radicals was assessed by **Akowuah *et al.*, (2005)**. After 30 minutes of incubation in the dark at 30°C, methyl alcohol was employed as a blank, and a UV-spectrophotometer was used to detect the absorbance at 517 nm after 30 minutes of incubation at 30°C in the dark. The antioxidant capacity for each sample was expressed as percent inhibition when measured in triplicate /each (an average value was calculated).

$$\% \text{ inhibition} = [(A_{control} - A_{sample})/A_{control}] \times 100$$

** A is the absorbance using an UV spectrophotometer at 517_{nm}.

Determination of total phenolic and flavonoid contents

The total polyphenols contents of fruits' juices and yogurt were measured using the "colorimetric method" of Folin–Ciocalteau according to **Singleton and Rossi, (1965**). Briefly, two grams of sample were grounded with methyl alcohol, sonicated for 10 min, filtrated, and then the volume was completed with methanol up to 50 ml. Only one ml of this solution was standing with half ml of Folin–Ciocalteu reagent for 3 min and then the reaction was stopped by 1.5 ml of Na2CO3, after half an hour at RT, the absorbance was determined by A UV/Vis spectrophotometer (HITACHI U900, Japan) as gallic acid mg/100g at 765 nm.

Also, the total flavonoids of the samples were determined calorimetrically using the aluminum chloride method according to **Ordonez *et al.*, (2006).** The total flavonoid content was measured by adding 500 µl of sample to 500 µl of the ethanoic solution of ammonium chloride 2%, and then the solution was left in darkness for an hour at room temperature and the absorbance was determined by A UV/Vis spectrophotometer (HITACHI U900, Japan), as quercetin (mg/100g sample).

HPLC analysis of yogurts' and juices' treatments

An Agilent 1260 series, Infinity, II LC, Waldbronn, Germany, was used for the HPLC analysis according to **Mradu et al., (2012).** The separation column specification is as follows: "4.6 mm x 250 mm i.d., 5 μm, Eclipse C18" which was kept at 40 °C during measurement. The used mobile phase of the apparatus is (A) water and (B) 0.05% trifluoroacetic acid dissolved in acetonitrile at a flow rate of 0.9 ml per min. The mobile phase was programmed sequentially as follows: 0 minutes (82% A), 0-5 minutes (80% A), 5-8 minutes (60% A), 8 minutes (12%), 12 minutes (15%), 15 minutes (16%), and 16 minutes (20%). At 280 nm, the wavelength detector was observed. For every one of the sample solutions, 5 l of injection volume was used.

Microbiological analysis:

The bacteria in the yogurt samples were cultured and counted according to **Terzaghi and Sandine (1975) ; Dave and Shah (1996), and Marshall, (1992)** for *Lactobacillus Bulgaricus, Streptococcus thermophilus* and *Bifidobacterium infantis*, in triplicate at 1, 7 and 14 days of the storage. The enumeration condition of each strain is indicated as follows: *Lactobacillus Bulgaricus*, MRS agar at 37 °C for 72 h in anaerobic condition, *Streptococcus thermophilus*, M17 agar at 37°C for 48 h in aerobic condition and *Bifidobacterium infantis* count was done using M17 agar at 30 °C for 72 h in anaerobic condition. The counting of these bacteria expressed as colony-forming units/ gram (log CFU/g) of each sample.

Statistical analysis:

All obtained data were subjected to statistical analysis using one-way analysis of variance (ANOVA) using the SPSS statistical software program version 19 (SPSS Inc., USA). Results were indicated by mean

± SD. The significant differences among treatments' means were explored according to Post Hoc Duncan's test at a 0.05 probability level using a statistical software package (InfoState) according to **Casanoves *et al.*, (2012)**.

Results and discussion

Physicochemical analysis of yogurt fortified with plum juice treatments:

The changes in pH and acidity of all yogurt treatments were investigated in this study and illustrated in Table (1). In general, it was found that pH values significantly declined during storage, and the opposite was for the TTA values. The lowest PH and highest acidity were observed in yogurt fortified with probiotic bacteria and plum juice (ranged from 4.29±0.03 to 3.11±0.04) and (0.93±0.01 to 1.05±0.01%), respectively compared to control yogurt (ranged from 4.69±0.02 to 3.98±0.03) and (0.84±0.01 to 0.96±0.02%), respectively during 14 days storage. The addition of plum juice to probiotic yogurt decreased the PH and increased acidity.

The decrease in pH may be due to starter culture bacteria, which are responsible for the post-acidification of yogurt (**Christopher *et al.*, 2009).** Starter bacteria of *L. bulgaricus* and *S. thermophilus* in cold conditions are responsible for the secretion of beta-galactosidase enzyme which converts lactose to lactic acid **(Christopher *et al.*, 2009).**

In this respect, **Al-Farsi and Lee, (2008)**, **and Meenakshi et al., (2018)** reported that lower pH and higher acidity values may be attributed to the acidity of the fruit because of the high content of phenolics, flavonoids, sugars and vitamins.

The effect of probiotic bacteria (*Bifidobacterium*) on both pH and acidity is contributed by the acidity caused by the regular starter culture, which is a critical factor for *Bifidobacterium* growth. So, a higher count of *Bifidobacterium* produces more metabolites, especially acetic acid, which lowers the pH and increases the acidity as well (**Sarvari et al., 2014 and Meenakshi et al., 2018**).

Table (1): **pH and acidity values of yogurt fortified with plum juice treatments**

Treatment	Storage period (d)	pH	Acidity (%)
Control yogurt	1	$4.69{\pm}0.02^{a}$	$0.84{\pm}0.01^{i}$
	7	$4.44{\pm}0.04^{c}$	$0.90{\pm}0.02^{gh}$
	14	$3.98{\pm}0.03^{h}$	$0.96{\pm}0.02^{cd}$
Yogurt fortified with plum juice	1	$4.38{\pm}0.04^{e}$	$0.92{\pm}\ 0.01^{ef}$
	7	$4.05{\pm}0.02^{g}$	$0.95{\pm}\ 0.03^{d}$
	14	$3.66{\pm}0.03^{j}$	$0.99{\pm}0.02^{b}$
Yogurt fortified with probiotic bacteria	1	$4.52{\pm}0.02^{b}$	$0.89{\pm}0.03^{h}$
	7	$4.41{\pm}0.02^{d}$	$0.91{\pm}0.04^{fg}$
	14	$3.89{\pm}\ 0.03^{i}$	$0.97{\pm}0.02^{c}$
Yogurt fortified with probiotic bacteria and plum juice	1	$4.29{\pm}0.03^{f}$	$0.93{\pm}0.01^{e}$
	7	$3.63{\pm}0.03^{k}$	$0.99{\pm}0.02^{b}$
	14	$3.11{\pm}0.04^{l}$	$1.05{\pm}0.01^{a}$

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$..

Data in Table (2) showed that the moisture content and T.S. were major constituents (80.44% and 18.67%, respectively) in juice, while fat, ash and protein were minor contents (0.04, 0.62 and 0.23%) respectively. Previous research by **Bozhkova, (2014**), reported that the total soluble solids of twenty-five types of plum ranged from 15.6 to 22.19%.

Also, the maximum moisture content was in yogurt fortified with probiotic bacteria and plum juice, and yogurt fortified with plum juice. Because plum juice contains a high level of moisture (80.44%), adding it to yogurt increases the moisture content. Moisture content, on the other hand, decreased significantly during storage. Recent research by **Rahman *et al.*, (2020),** found that the addition of strawberry juice to yogurt caused the moisture content to increase.

Also, as shown in Table (2) and Figure (1), yogurt fortified with probiotic bacteria and 10% plum juice increased total solids (T.S.) (13.73±0.02%) significantly more than yogurt fortified with plum juice without probiotics (13.69±0.03%), followed by probiotic yogurt without juice (13.62±0.03%), compared to control yogurt (13.53±0.02%). The results showed that the addition of fruit juice to yogurt increased the total solid (T.S). These results may be due to plum juice being rich in mineral salts, pectin and another carbohydrate like mono and disaccharides. **(Yagmur and Taskin, 2011)**, reported that plums have higher amounts of carbohydrates and minerals.

The T.S. content of probiotic yogurt, on the other hand, was higher (ranging from 13.62± 0.03% to 13.88± 0.07%) than the T.S. content of control yogurt (which ranged from 13.53± 0.02 to 13.83± 0.06%). The increase in T.S. was observed during the storage period for all treatments. The increase in T.S. may be due to a decrease in yogurt moisture percentage during storage **(Pagthinathan *et al.*, 2018).**

By comparing the yogurt treatments, it indicated that there are no significant differences in fat and protein content between the control yogurt and the probiotic yogurt. Meanwhile, plum juice-fortified yogurt had a lower fat and protein content than yogurt without juice. The addition of plum juice to yogurt decreases fat and protein because of the low content of fat and protein in the juice which leads to a decrease in fat and protein. Recent research by **Ismail *et al.*, (2020),** found that the addition of fruit juices to milk decreases its protein content.

In general, all the chemical composition parameters of, T.S., fat, protein and ash percent are gradually increasing with time of storage, these results are similar to those reported by **Hernández-Herrero and Frutos, (2014); Christopher et al., (2009); Hallim et al., (2019), and Ismail et al., (2020).**

These findings could be explained by the fact that during yogurt fermentation with a long storage time of up to two weeks, the pH gradually decreased and the syneresis was elevated, causing the yogurt to lose moisture and all previous parameters to rise over time. Moreover, the T.S., protein, fat and ash % in 100g of fresh plum juice were 18.67, 0.62, 0.04 and 0.23%, respectively.

According to **Hassanein et al., (2014),** fortifying yogurt with fruit juice may lower its chemical composition parameters due to the fruit's high moisture and low fat and protein contents, and these treatments cause syneresis to increase by storage and moisture loss by evaporation, resulting in an increase in the percentage of the yogurt's chemical parameters.

Table (2): Chemical composition of yogurt treatments fortified with plum juice

Chemical composition	Plum juice	Storage period (d)	Control yogurt	Yogurt fortified with plum juice	Probiotic yogurt	Probiotic yogurt fortified with plum juice
Total solid (%)	18.67	1	13.53±0.02[h]	13.69±0.03[f]	13.62±0.03[g]	13.73±0.02[e]
		7	13.61±0.02[g]	13.76±0.03[e]	13.69±0.03[f]	13.75±0.03[e]
		14	13.82±0.06[d]	13.98±0.08[b]	13.88±0.07[c]	14.02±0.04[a]
Moisture (%)	80.44	1	78.52±0.02[a]	78.85±0.03[de]	78.42±0.03[b]	78.81±0.02[ef]
		7	78.17±0.02[b]	78.57±0.03[f]	78.06±0.03[d]	78.49±0.03[j]
		14	77.61±0.06[g]	78.03±0.08[h]	77.54±0.07[g]	77.91±0.04[h]
Fat (%)	0.04	1	3.29±0.09[bcd]	3.03±0.07[d]	3.27±0.08[bcd]	3.00±0.09[e]
		7	3.48±0.05[ab]	3.16±0.11[cd]	3.48±0.08[ab]	3.19±0.10[bcd]
		14	3.65±0.03[a]	3.39±0.10[abc]	3.66±0.07[a]	3.42±0.08[abc]
Protein (%)	0.62	1	3.74±0.03[d]	3.50±0.02[h]	3.77±0.02[cd]	3.53±0.02[h]
		7	3.81±0.05[bc]	3.58±0.03[g]	3.85±0.02[b]	3.62±0.03[fg]
		14	3.96±0.06[a]	3.64±0.09[ef]	3.95±0.08[a]	3.67±0.06[e]
Ash (%)	0.23	1	0.92±0.03[d]	0.93±0.03[d]	0.92±0.03[d]	0.93±0.03[d]
		7	0.93±0.02[d]	0.95±0.03[c]	0.92±0.02[d]	0.95±0.04[c]
		14	0.96±0.04[bc]	0.96±0.02[bc]	0.97±0.04[ab]	0.98±0.03[a]

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$.

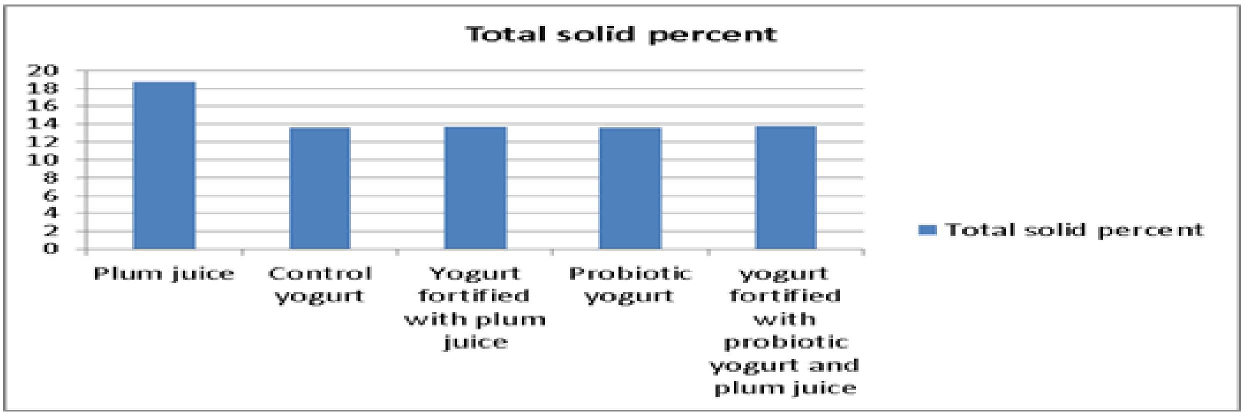

Figure (1): Total solid content different yogurt treatment

Rheological properties of yogurt fortified with plum juice treatments:

The loss of liquid from yogurt during the storage period is known as syneresis and is used as an indicator or a key to the quality of the yogurt itself **(Vital *et al.*, 2015, and Dabija *et al.*, 2018)**. Results in Table (3) showed that the syneresis increased and viscosity decreased gradually during the storage period. The lowest whey syneresis was for control yogurt ranging from 6.6 ml on day 1 up to 11.9 ml while the lowest viscosity was yogurt fortified with plum juice ranging from 151±12 cP) to 75±9 on (cP) day 14. The results revealed that there are no significant differences in syneresis value and viscosity between control yogurt and probiotic yogurt. Also, no significant difference was observed between yogurts fortified with plum juice and probiotic yogurt fortified with plum juice.

However, the addition of 10 percent plum juice to the previous treatment caused the highest syneresis amount and lowest viscosity of all treatments, which varied from 16.3 ml at day 1 up to 22.2 ml on 14 days and from 149±14 cP on day 1 to 70±9 cP at day 14. Many studies had indicated that the syneresis amount is increasing with storage progress due to yogurt acidity development causing the water holding capacity to be reduced during storage progress (**Silva *et al.*, 2017 and Ismail *et al.*, 2020). Saleh *et al.*, (2018)** confirmed these observations, finding that the viscosity of yogurt was decreased during storage time (15 days).

Furthermore, many studies have shown that when storage increases syneresis, viscosity decreases **(Hassanein et al., 2014; Hallim et al., 2019, and Hassaan et al., 2019).** Probiotic yogurt fortified with plum juice showed the highest syneresis values compared with control yogurt. However, probiotic yogurt fortified with plum juice, showed an improvement in both parameters. These value changes during storage may be related to the increased changes in casein particle rearrangement (protein-protein pound) caused by the acidity fermentation of yogurt **(Hassanein et al., 2014; Shahbandari et al., 2016).**

Table (3): Syneresis and viscosity values of yogurt treatments fortified with plum juice

Parameters	Storage period (d)	Control yogurt	Yogurt fortified with plum juice	Probiotic yogurt	Probiotic yogurt fortified with plum juice
Syneresis (ml)	1	6.6±0.7[g]	16.3±0.8[d]	6.6±0.7[g]	16.2±0.9[d]
	7	9.6±0.6[f]	18.4±0.9[c]	9.4±0.5[f]	18.3±0.8[c]
	14	11.9±0.7[e]	22.2±1.5[a]	11.9±0.9[e]	21.5±1.4[b]
Viscosity (cP)	1	163±11[a]	149±14[b]	165±15[a]	151±12b
	7	137±14[c]	116±13[d]	143±12[b]	120±9[cd]
	14	88±9[e]	70±9[f]	89±8[e]	75±9[f]

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$.

Sensory evaluation of yogurt fortified with plum juice treatments:

Results in Table (4), showed that all the sensory properties indicators gradually declined with a storage period of up to 14 days, these results are in accordance with **Christopher *et al.*, (2009); Hallim *et al.*, (2019); Atwaa *et al.*, (2020), and Ismail *et al.*, (2020).** Yogurt fortified with plum juice gained the lowest scores for body and texture percent; the present results are shown in Figure (2). Yogurt fortified with plum juice had a lower pH and higher acidity values than regular yogurt. This may be attributed to the sugars in the fruit encouraging the lactic acid bacteria to transform sugar into lactic acid, acetaldehyde, diacetyl, and formic acid **(Meenakshi et al., 2018).** The accumulation of all these fermentation products corresponds to an increase in acid levels, which affects the texture of the yogurt.

On the other hand, there is no significant difference P≤0.05 observed in appearance between control yogurt and probiotic yogurt fortified with plum juice at one day and 14 day storage.

Table (4): Organoleptic evaluation of yogurt treatments fortified with plum juice

Parameters	Storage period (d)	Control yogurt	Yogurt fortified with plum juice	Probiotic yogurt	Probiotic yogurt fortified with plum juice
Body and texture (30)	1	29±0.25[a]	27±0.26[f]	28.1±0.23[c]	28.5±0.22[b]
	7	28±0.20[d]	25±0.21[h]	26.8±0.23[g]	27.2±0.23[e]
	14	25±0.19[h]	19±0.22[j]	22.5±0.20[l]	23±0.22[i]
Acidity (10)	1	9.5±0.20[a]	9±0.19[c]	9.2±0.20[b]	9.2±0.18[b]
	7	8.1±0.21[h]	8.5±0.15[e]	8.7±0.17[d]	8.0±0.17[i]
	14	8.5±0.18[e]	8.3±0.18[g]	8.5±0.16[e]	8.4±0.15[f]
Appearance (15)	1	14.3±0.15[a]	13±0.21[d]	13.8±0.21[a]	14.2±0.19[a]
	7	13.8±0.17[b]	12±0.15[e]	13±0.19[d]	13.4±0.18[c]
	14	12.1±0.22[e]	10.5±0.23[f]	12±0.21[e]	12.0±0.24[e]
Flavor (45)	1	44.1±0.26[a]	42±0.22[f]	42.1±0.24[e]	42.5±0.21[d]
	7	43.2±0.21[b]	40.5±0.21[g]	42.5±0.20[d]	43±0.19[c]
	14	40.4±0.22[h]	38±0.24[k]	39±0.23[j]	40±0.20[i]
Overall acceptability (100)	1	96.9±0.86[a]	91±0.88[e]	93.2±0.88[c]	94.1±1.10[b]
	7	94.1±0.21[b]	86±0.72[f]	91±0.79[e]	91.6±0.77[d]
	14	86±0.81[f]	75.8±0.87[i]	82±0.81[h]	83.4±0.81[g]

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at p ≤ 0.05.

However, the fortification of yogurt with plum juice affects the overall acceptability of the yogurt. As well, probiotic yogurt fortified with plum juice had nearly the same overall acceptability as the control yogurt. On the other hand, the storage period influenced the overall acceptability of all yogurt samples. According to **Coggins et al., (2010)** yogurt sensory evaluation declines during the storage period. Pervious results by **Sallm et al., (2010**) found that the fresh juice had significant impact on yogurt acceptability both before and after storage.

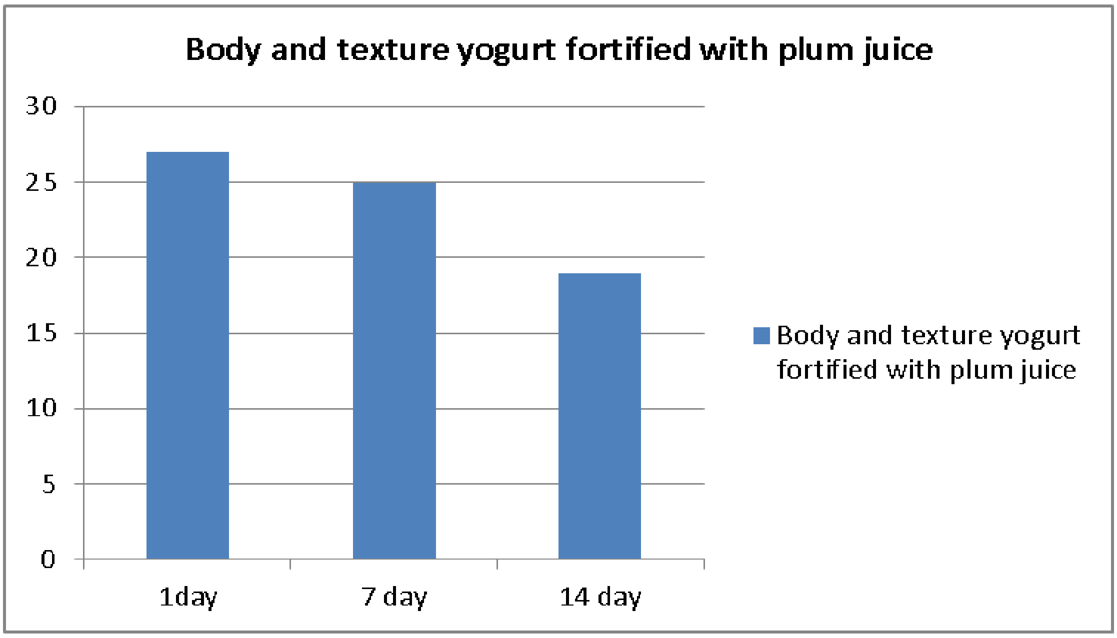

Figure (2): Body and texture of yogurt fortified with plum juice during storage period

Chemical analysis

Determination of total phenolic, flavonoids contents and antioxidant activity of yogurt fortified with plum juice treatments:

The total phenolic, total flavonoid contents and antioxidant activity of plum juice, control yogurt, yogurt fortified with plum juice, and probiotic yogurt fortified with plum juice are presented in Table (5). The total phenolic, total flavonoid contents and antioxidant activity of plum juice were 108.782±0.21 mg/100g, 5.776±0.09 mg/100g and 91.97±0.13 %, respectively. It is well known that the phenolic and flavonoid contents of fruits are responsible for their antioxidant activity, which is highly correlated to each other and plum fruits have a high content of phenolic and flavonoid compounds such as anthocyanin **(Perez-Fons *et al.*, 2010; Cosmulescu, *et al.*, 2015, and Akbar, 2019).**

Data in Table (5) and Figure (3) revealed that total phenol in probiotic yogurt fortified with plum juice and yogurt fortified with plum juice was higher than in control yogurt (11.46±1.18 and 11.952±1.58 mg/g, respectively), The high phenolic content of plum juice (108.782±0.21 mg/100g) could explain the increase in total phenol in all plum yogurt treatments. These results are in agreement with **Blassy et al., (2020).**

Yogurt fortified with plum juice during storage time of 1, 7 and 14 days showed total phenolic content of 20.418±1.92, 15.645±1.52 and 13.649±1.03 as GA (mg/100 g sample), respectively, and this treatment during storage time of 1, 7 and 14 days showed total flavonoid content of 2.698±0.15, 2.236±0.25 and 1.864±0.19 as Catechin (mg/100 g sample), respectively. In addition, this treatment during storage time of 1, 7 and 14 days showed antioxidant activity as DPPH inhibition percent of 95.60 ±0.76, 88.38±1.33 and 79.31±0.56 %, respectively. Previous research by **Cosmulescu et al., (2015)** reported that the whole plum fruit and pulp has a higher content of natural phenolic antioxidant.

However, probiotic yogurt showed higher total flavonoid content and antioxidant activity values than control yogurt. Furthermore, there was no significant difference ($P > 0.05$) in phenolic compounds and flavonoids between probiotic yogurt fortified with plum juice and yogurt fortified with plum juice, but in terms of antioxidant activity, there was a significant difference $P \leq 0.05$ between probiotic yogurt fortified with plum juice and yogurt fortified with plum juice during storage time 1 and 7 days.

Bifidobacterium may include certain possible bioactive substances that act as electron donors and successfully react with free radicals to transform them into more stable molecules and put an end to the radical chain reaction, which may be the reason for the elevation of the antioxidant activity **(Vitheejongjaroen *et al.*, 2021)**.

On the other hand, probiotic yogurt fortified with plum juice during storage time of 1, 7 and 14 days showed antioxidant activity as DPPH inhibition percent of 98.73±0.65, 89.84±1.94 and 80.19±0.97 %, respectively. So, the addition of fruit juice to yogurt led to an increase in total phenolic content, total flavonoid content and antioxidant activity of the treatments when compared to the control.

Table (5): Total phenolic content, total flavonoid content and antioxidant activity of yogurt treatments fortified with plum juice

Parameters	Plum juice	Storage period (d)	Control yogurt	Yogurt fortified with plum juice	Probiotic yogurt	Probiotic yogurt fortified with plum juice
Total phenolic content (mg/100g sample)	108.782 ±0.21	1	11.464±1.18[d]	20.418±1.92[a]	11.952±1.85[d]	20.600±1.13[a]
		7	9.122±1.13[e]	15.645±1.52[b]	9.338±1.24[e]	15.649±1.32[b]
		14	2.631±1.17[f]	13.649±1.03[c]	2.873±1.46[f]	13.754±1.34[c]
Total flavonoid content (mg/100g sample)	5.776 ±0.09	1	1.607±0.13[e]	2.698±0.15[a]	1.952±0.16[d]	2.745 ±0.19[a]
		7	1.371±0.17[g]	2.236±0.25[c]	1.559±0.28[f]	2.354±0.22[b]
		14	1.245±0.14[h]	1.864±0.19[e]	1.398±0.16[f]	1.922±0.18d[e]
Antioxidant activity as DPPH inhibition (%)	91.97 ±0.13	1	46.44±0.92[g]	95.60 ±0.76[b]	50.31±0.27[f]	98.73±0.65[a]
		7	38.76±1.40[h]	88.38±1.33[d]	38.93±1.63[h]	89.84±1.94[c]
		14	20.18±0.69[i]	79.31±0.56[e]	20.79±0.92[i]	80.19±0.97[e]

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$.

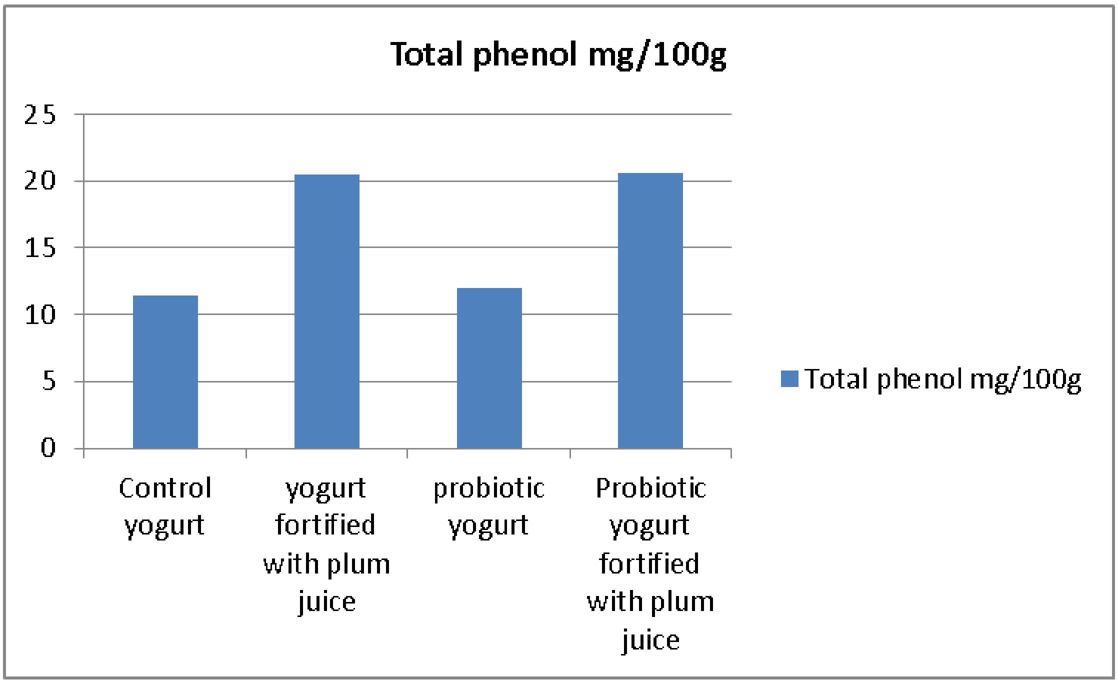

Figure (3): Total phenolic compounds of different treatments

Fortification of yogurt and other milk products with fruits has increased their phenolic, flavonoid contents and antioxidant activity **(El-Samahy et al., 2014; Kumar and Kumar, 2016; Bchir et al., 2019; Hallim et al., 2019, and Atwaa et al., 2020).**

In general, the results from Table (5): showed that total phenolic, flavonoid contents and antioxidant activity values gradually decreased along with the storage for all treatments. This may be a result of the oxidation of phenolic compounds or its reaction with caseins. Also, the presence of lactic acid, which is secreted by the cultural bacteria causes, phenolic compounds to be decomposed as well **(Dalling, 1986; Cho *et al.*, 2017 and Han *et al.*, 2019).**

HPLC analysis of phenolic compounds in yogurt treatments

The amount of phenolic compounds in plum juice, control yogurt, probiotic yogurt, and probiotic yogurt fortified with plum juice are presented in Figures (4-8) and Table (6). Twelve phenolic compounds were detected in plum juice, the majority of them ellagic acid (28.37±0.01µg/ml), catechin (12.49±0.01µg/ml), methyl gallate (8.04±0.01µg/ml), naringinin (6.69±0.01µg/ml) and chlorogenic acid (3.29±0.010µg/ml) while, syringic acid was not detected. Different results obtained by **Jaiswal *et al*., (2013),** reported that the main phenolic compounds in plum fruit were chlorogenic acid and proanthocyanidins. Other research by **Celik *et al*., (2017),** indicated that chlrogenic acid was a major phenolic compound in plum fruit.

The main phenolic compound in control yogurt was gallic acid (0.87±0.01 µg/ml), while quercetin (1.26±0.01 µg/ml) was present in yogurt fortified with plum juice and catechin (1.08±0.01 µg/ml) was present in probiotic yogurt fortified with plum juice. However, the lowest concentration was of vanillin in plum juice (0.14±0.01 µg/ml), naringenin in control yogurt (0.13±0.01 µg/ml) and probiotic yogurt (0.40±0.01 µg/ml), methyl gallate in yogurt fortified with plum juice (0.06±0.01 µg/ml) and probiotic yogurt fortified with plum juice (0.05±0.01 µg/ml), respectively.

The most plentiful phenolic compounds in probiotic yogurt fortified with plum juice were catechin, gallic acid, and chlorogenic acid followed by quercetin, ellagic acid, naringenin, methyl gallate and ferulic acid. Pyro catechol, rutin, vanillin, syringic acid and daidzein were not detected. Recent research by **Johnson *et al*., (2022)** found that the major phenolic compounds in plum flesh were cynadinin -3-glucoside, followed by ellagic acid, gallic acid, catechin, myricetin and chlorogenic acid while naringenin was a minor component. From these results, it can be observed that the yogurt fortification with probiotics and plum juice increased the phenolic compounds content in yogurt. **Taneva and Zlatin (2020)** reported that the addition of fruit to yogurt increases phenolic content.

Table (6): HPLC analysis of phenolic compounds in yogurt treatments fortified with plum juice

Phenolic components	Conc. (µg/ml)				
	Plum juice	Control yogurt	Probiotic yogurt	Yogurt fortified with plum juice	Probiotic yogurt fortified with plum juice
Gallic acid	2.90±0.01^{c}	0.87±0.02^{a}	0.99±0.03^{b}	0.87±0.02^{d}	1.03±0.02^{e}
Chlorogenic acid	3.29±0.02^{c}	0.42±0.03^{a}	1.55±0.02^{b}	0.17±0.01^{d}	0.64±0.04^{e}
Catechin	12.49±0.01^{a}	ND	ND	0.33±0.01^{b}	1.08±0.02^{c}
Methyl gallate	8.04±0.01^{a}	ND	ND	0.06±0.01^{b}	0.05±0.03^{c}
Pyro catechol	0.47±0.03^{a}	ND	ND	ND	ND
Rutin	0.44±0.02^{a}	ND	ND	0.07±0.02^{b}	ND
Vanillin	0.14±0.01^{a}	ND	ND	ND	ND
Syringic acid	ND	0.17±0.01^{a}	ND	0.23±0.01^{b}	ND
Ellagic acid	28.37±0.01^{a}	ND	ND	ND	0.46±0.01^{b}
Ferulic acid	0.50±0.03^{a}	ND	ND	ND	0.03±0.02^{b}
Naringenin	6.69±0.02^{c}	0.13±0.01^{a}	0.40±0.02^{b}	0.10±0.02^{d}	0.11±0.03^{d}
Daidzein	0.25±0.01^{b}	0.18±0.01^{a}	ND	0.13±0.01^{c}	ND
Querectin	1.38±0.02^{a}	ND	ND	1.26±0.01^{b}	0.51±0.03^{c}

ND; not detected, The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$.

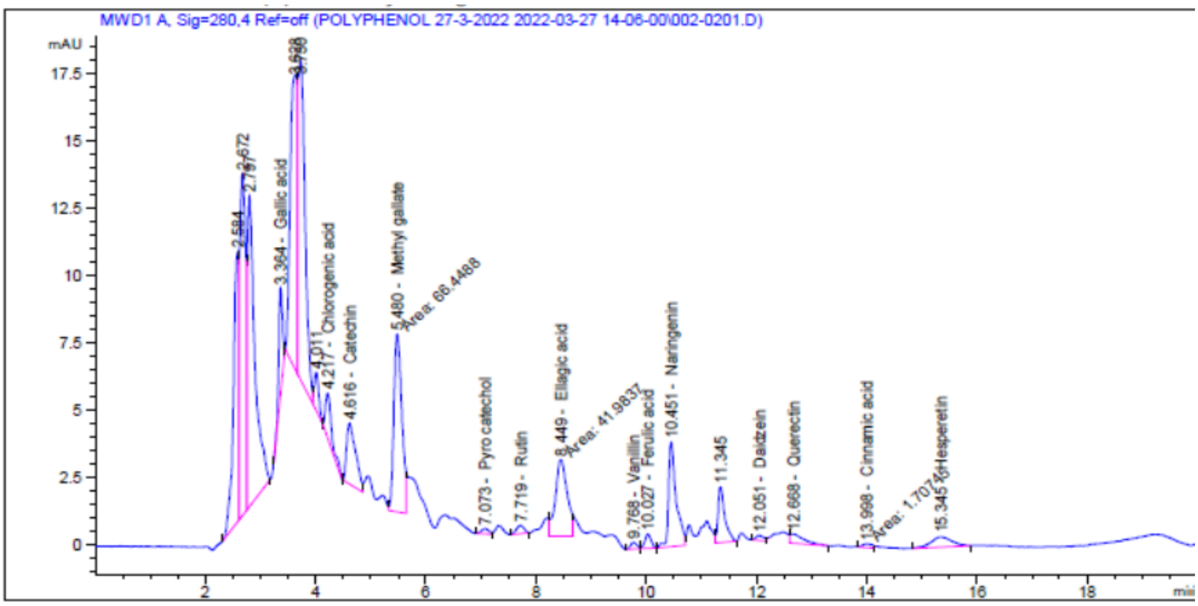

Figure (4): HPLC chromatogram of phenolic contents of plum juice

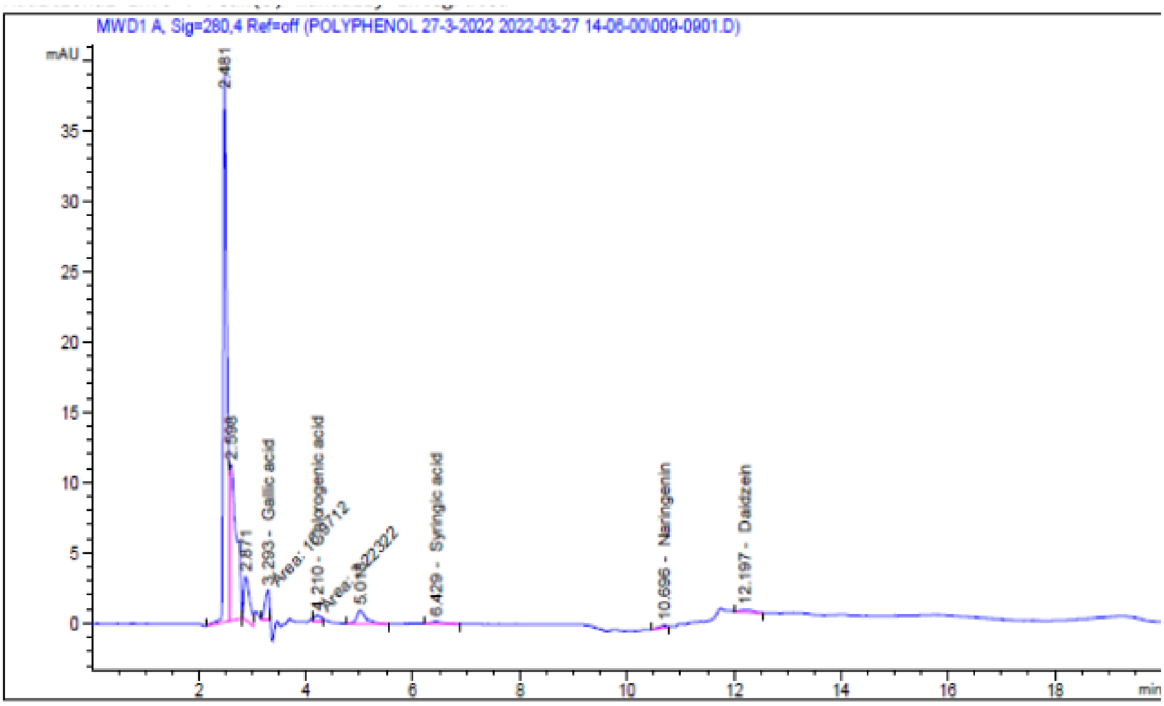

Fig (5): HPLC chromatogram of phenolic contents of control yogurt

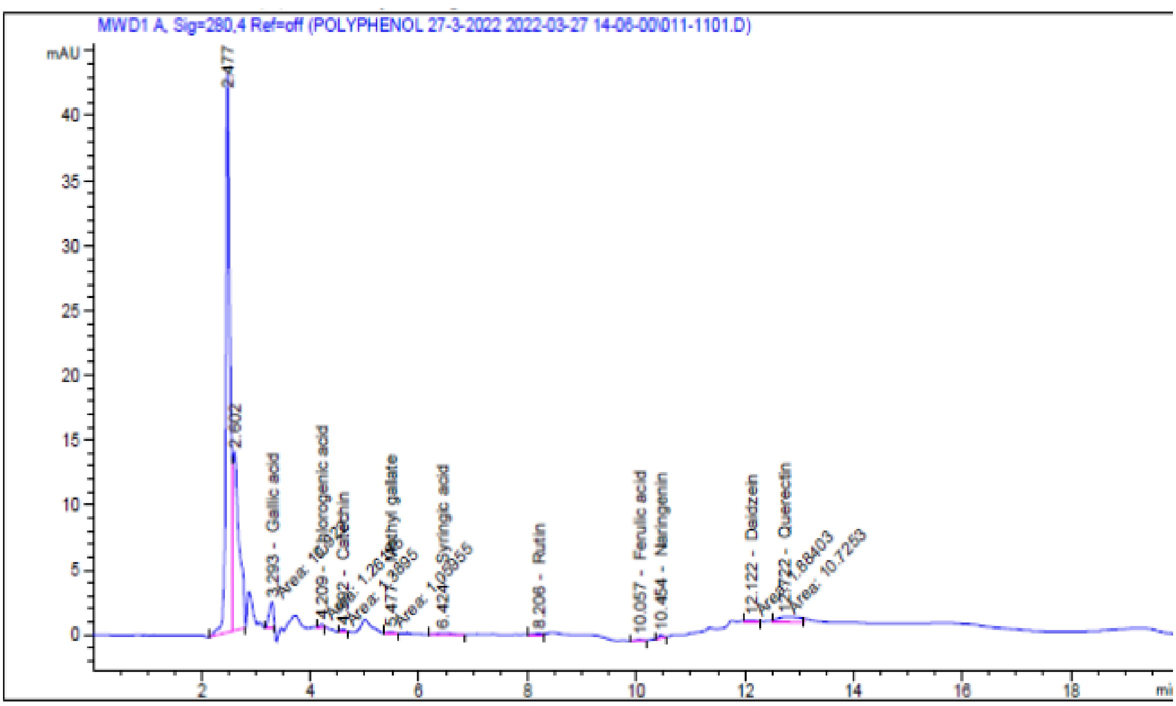

Fig (6): HPLC chromatogram of phenolic contents of yogurt fortified with plum juice

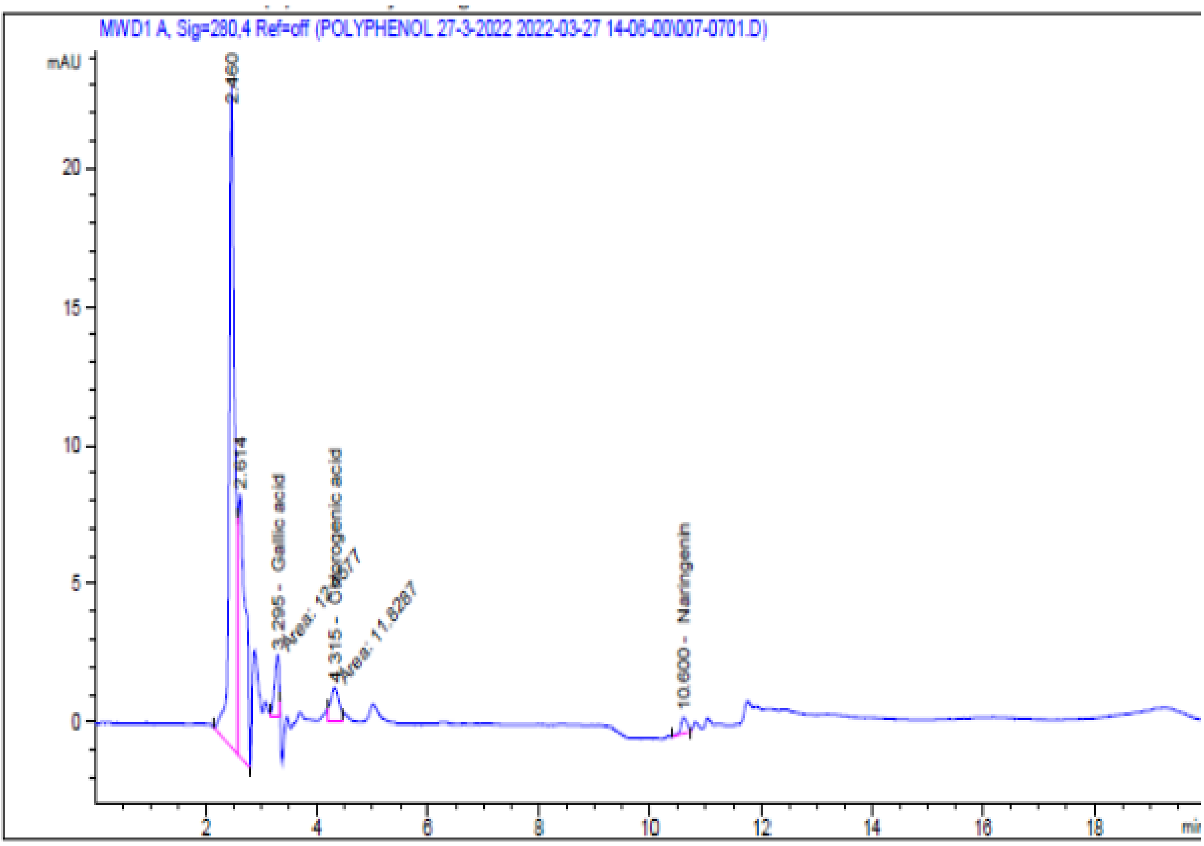

Fig (7): HPLC chromatogram of phenolic contents of probiotic yogurt

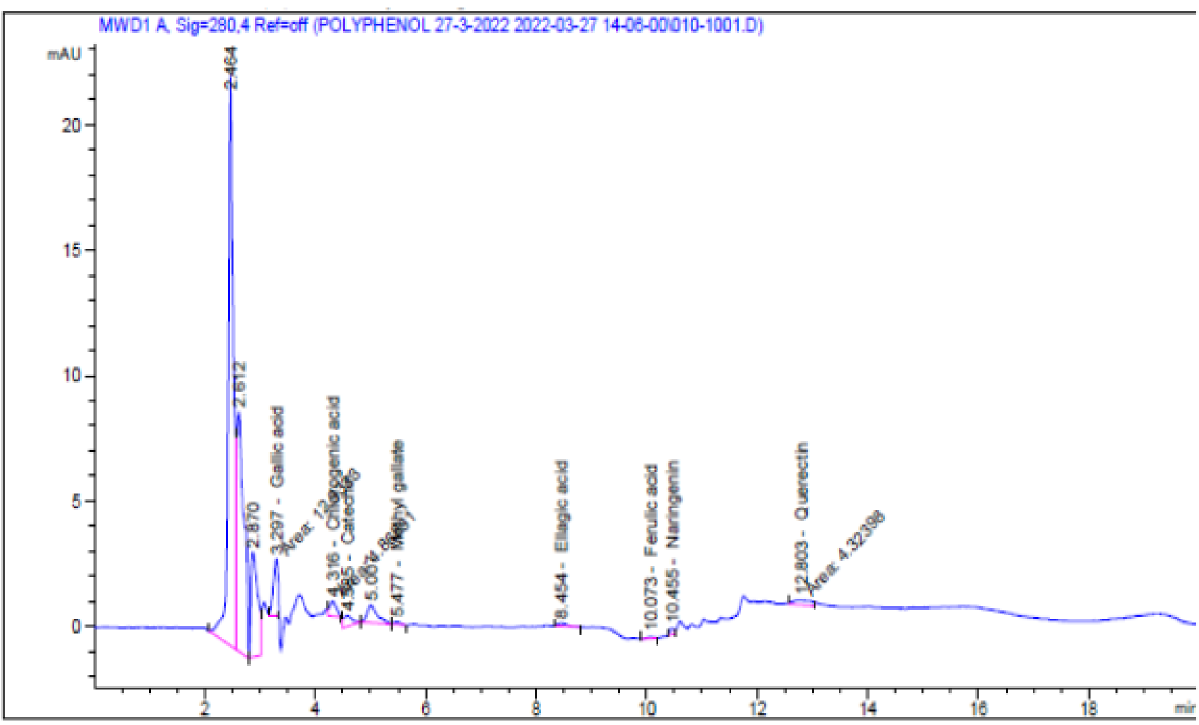

Fig (8): HPLC chromatogram of phenolic contents of probiotic yogurt fortified with plum juice

Microbiological examination of yogurt fortified with plum juice.

Tables 7 and 8 show that the total viable count of yogurt fortified with plum juice in the presence or absence of *Bifidobacterium* is indicated as (CFU/g). The *Bifidobacterium* count was the highest of all total bacterial counts across all treatments, according to the results.

However, the bacterial count of the probiotic yogurt fortified with 10% plum is lower than the bacterial count of probiotic yogurt, these results are in line with **Christopher *et al.*, (2009); Hallim *et al.*, (2019), and Atwaa *et al.*, (2020).**

Besides, *S. thermophilus* and *L. bulgaricus* numbers declined during the storage period. These results are consistent with the finding of **Buriti et al., (2007**), where the lactic acid bacteria and acidity of fruits may negatively affect the growth and viability of starter bacteria in dairy products.). In general, the increase in *Bifidobacterium* counts may be due to these bacteria's anaerobic preference; with a long storage period, the oxygen percent in yogurt decreases (**Tripathi and Giri, 2014)**. However, it was stated that acidity is a main factor for *Bifidobacterium* growth, and the acidity of the fruit and starter culture metabolism causes a reduction in the viable count of *Bifidobacterium* during long storage (**Dave and Shah, 1998 and Meenakshi et al., 2018).**

Furthermore, the fortification of plum juice yogurt, with or without *Bifidobacterium*, had a lower bacterial count than without it. This is because plum juice has a high concentration of phenolic compounds and flavonoid compounds, resulting in a low bacterial count **(Meenakshi et al., 2018)**.

Table (7): Viable counts of lactic acid bacteria (CFU/g) of yogurt fortified with plum juice during storage at 4°C for 14 days

Treatment	Storage period (d)	*Lactobacillus*	*Streptococcus*
		CFU×10^6/g	
Control yogurt	1	8.46±0.02^b	8.50±0.02^a
	7	8.34±0.04^d	8.38±0.03^c
	14	6.73±0.01^j	7.61±0.01^e
Yogurt fortified with plum	1	7.43±0.02^g	7.50±0.02^f
	7	7.24±0.03^i	7.39±0.04^h
	14	5.16±0.02^l	6.53±0.01^k

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$

.

Table (8): Viable counts of lactic acid bacteria and *Bifidobacterium* (CFU/g) of yogurt fortified with plum juice during storage at 4°C for 14 days

Treatment	Storage period (d)	*Lactobacillus*	*Streptococcus*	*Bifidobacterium*
		CFU×10^6/g		
Probiotic yogurt	1	8.55±0.01^c	8.54±0.02^c	8.89±0.01^a
	7	8.38±0.03^e	8.41±0.04^d	8.63±0.02^b
	14	6.77±0.01^o	7.70±0.02^h	8.11±0.01^f
Probiotic yogurt fortified with plum	1	7.48±0.02^k	7.59±0.02^j	7.75±0.02^g
	7	7.29±0.03^m	7.33±0.04^l	7.61±0.02^i
	14	5.35±0.01^q	6.61±0.03^p	6.92±0.01^n

The means in each column with similar superscript (s) are non- significantly different while different letters are significantly different at $p \leq 0.05$

Conclusion

Yogurt is counted as one of the most favorable and demandable dairy products all over the world. The aim of this work is to investigate both the fortification of yogurt with plum juice and culturing it with *Bifidobacterium* as a probiotic. The results indicated that culturing yogurt with *Bifidobacterium* along with regular starters has no different sensory properties than regular yogurt. However, only the addition of plum juice to yogurt has enhanced its total phenolic, total flavonoid contents and antioxidant effects. However, this resulted in lower body and texture properties, which could be compensated for by adding nonfat dry milk. When yogurt is cultured with *Bifidobacterium* alongside regular starters and fortified with 10% plum juice, its total phenolic and total flavonoid contents, antioxidant effects, and sensory properties improve. So, the enrichment of yogurt with plum juice and *Bifidobacterium* gives more variable choices to the consumers, which has many influences including health benefits, enriched nutrients and economic impacts.

REFERENCES

Abdelshafy, A. M., Belwal, T., Liang, Z., Wang, L., Li, D., Luo, Z. and Li, L. (2021): A comprehensive review on phenolic compounds from edible mushrooms: occurrence, biological activity, application and future prospective. Critical Reviews in Food Science and Nutrition, 62 (22): 6204-6224.

A.O.A.C. (1995): Official methods of analysis 16th Edition. The Association of Official Analytical Chemists. AOAC International Publisher, Washington DC, USA.

A.O.A.C. (2000): Official Methods of Analysis. 17th Edition. The Association of Official Analytical Chemists. AOAC International Publisher, Gaithersburg, MD, USA.

Ahmad, I., Hao, M., Li, Y., Zhang, J., Ding, Y. and Lyu, F. (2022): Fortification of yogurt with bioactive functional foods and ingredients and associated challenges - A review. Trends in Food Science and Technology, 129 (11): 558-580.

Akan, K., Yerlikaya, O., Bayram, O. Y. and Kinik, O. (2022): The effect of aqueous extracts of some plants on in vitro antioxidant and antidiabetic activity of probiotic yogurt. Journal of Food Science and Technology, 59 (9): 3359-3366.

Akbar, S. A. (2019): Evaluation of antioxidant activity on plum fruit (*Prunus domestica* L.*)* Skin extract applied for natural acid-base indicator. Journal of Islamic Science and Technology, 5 (1): 21-29.

Akowuah, G., Ismail, Z., Norhayati, I. and Sadikun, A. (2005): The effects of different extraction solvents of varying polarities of polyphenols of Orthosiphon stamineus and evaluation of the free radical-scavenging activity. Food Chemistry, 93(2): 311-317.

Al-Farsi, M. A. and C.Y. Lee, (2008): Nutritional and functional properties of date. A review. Critical Reviews in Food Science and Nutrition, 48(10): 877-887.

Aryana, K. J. (2003): Folic acid fortified fat free plain set yogurts. International Journal of Dairy Science and Technology, 56 (4): 219-222.

Atwaa, E. H., Abou Sayed-Ahmed, E. A. and Hassan, M. A. A. (2020): Physicochemical, microbiological and sensory properties of low-fat probiotic yogurt fortified with mango pulp fiber waste as source of dietary fiber. Journal of Food and Dairy Science, Mansoura University, 11 (9): 271-276.

Bchir, B., Bouaziz, M., Blecker, C. and Attia, H. (2019): Physico-Chemical, antioxidant activities, textural, and sensory properties of yogurt fortified with different states and rates of pomegranate seeds (*Punica granatum* L.): Journal of Texture Studies, 51 (3): 475-487.

Blassy, K., Osman, M., Gouda, A. and Hamed, M. (2020): Functional properties of yogurt fortified with fruits pulp**.** Ismailia Journal of Dairy Science, 7 (1): 1-9.

Bondam, A. F., Da-Silveira, D. D., Dos-Santos, J. P. and Hoffmann, J. F. (2022): Phenolic compounds from coffee by-products: Extraction and application in the food and pharmaceutical industries. Trends in Food Science and Technology, 123 (8): 172–186.

Bozhkova, V. (2014): Chemical composition and sensory evaluation of plum. Trakya University Journal of Natural Sciences, 15(1): 31-35.

Buriti, f., Komatsu, T. and Saad, S. (2007): Activity of passion fruit (*Passiflora edulis*) and guava (*Psidium guajava*) pulps on *Lactobacillus acidophilus* in refrigerated mousses. Brazilian Journal of Microbiology, 38 (2): 315-317.

Casanoves, F., Balzarini, M., Di-Rienzo, J., Gonzalez, L., Tablada M., Robledo, C. W. (2012): infoStat. User Manual, Cordoba, Argentina.

Celik, F., Gundogdu, M., Alp, S., Muradoglu, F., Ercisli, S., Gecer, M. K. and Canan, I. (2017): Determination of phenolic compounds, antioxidant capacity and organic acids contents of *Prunus omestica L*., *Prunus cerasifera Ehrh*. and *Prunus spinosa L*. fruits by HPLC. Acta Chromatographica, 29 (4): 507-510**.**

Chavan J., Jagtap U., Gaikwad N., Dixit G. and Bapat V. (2013): Total phenolics, flavonoids and antioxidant activity of Saptarangi (*Salacia chinensis* L.) fruit pulp. Journal of Plant Biochemistry and Biotechnology, 22 (4): 409-413.

Cho, W., Yeon, S., Hong, G., Kim, J., Tsend-Ayush, C. and Lee, C. (2017): Antioxidant activity and quality characteristics of yogurt added green olive powder during storage. Korean Society for Food Science of Animal Resources, 37 (6): 865-872.

Christopher, M. D., Reddy, V. P. and Venkateswarlu, K. (2009): Viability during storage of two Bifidobacterium bifidum strains in set and stirred flavored yogurts containing whey protein concentrates. Natural Product Radiance, 8 (1): 25-31.

Cosmulescu, S., Trandafir, I., Nour, V. and Botu, M. (2015): Total phenolic, flavonoid distribution and antioxidant capacity in skin, pulp and fruit extracts of plum cultivars. Journal of Food Biochemistry, 39 (1): 64-69.

Dabija, A., Codină, G., Ropciuc, S., Gâtlan, A. and Rusu, L. (2018): Assessment of the antioxidant activity and quality attributes of yogurt enhanced with wild herbs extracts. Journal of Food Quality, 8 (6): 1-12.

Dalling, M. J. (1986): Plant proteolytic enzymes. CRC Press, 1st ed., FL, USA, (PP) 475-530.

Dave, R. I. and Shah, N. P. (1996): Evaluation of media for selective enumeration of *Streptococcus thermophiles, Lactobacillus delbrueckii ssp. bulgaricus, Lactobacillus acidophilus and Bifidobacterium ssp.* Journal of Dairy Science, 79 (9): 1529-1536.

Dave, R. I. and Shah, N.P. (1998): Ingredient supplementation effects on viability of probiotic bacteria in yogurt. Journal of Dairy Science, 81 (11): 2804-2816.

El-Samahy, S. K., Gaballah, A. A, Embaby, H. E., Hamed, Y. S. and Khalil, R. A. (2014): Improving the nutritional value and quality of yogurt drink by using Cactus pear (*Opuntia dillenii*) pulp. Egyptian Journal of Food Science, 42 (2): 133-144.

Granato, D., Barba, F. J., Kovačević, D. B., Lorenzo, J. M., Cruz, A. G. and Putnik, P. (2020): Functional Food**:** Products Development**,** Technology Trends, Efficacy Testing, and Safety. Annual Review, Food Science and Technology, 11 (1): 93-118.

Gurnani, N., Mehta, D., Gupta, M. and Mehta, B. K. (2014): Natural products: source of potential drugs. African Journal of Basic and Applied Sciences, 6 (6): 171-186.

Hallim, A. M., Rabie, A., El-Shewey, M. A. and Abdel-Ghany, A. S. (2019): Evaluation of physicochemical properties and antioxidant activity of stirred yogurt fortified with pomegranate and cactus pear juices. Zagazig Journal of Agriculture Research, 46 (6): 1995-2008.

Han, J., Chang, Y., Britten, M. and Lacroix, M. (2019): Interactions of phenolic compounds with milk proteins. European Food Research and Technology, 245 (9): 1881-1888.

Hassaan, M. G., Ahmed, R. B. and Khider, M. K. (2019): Low Fat Flavored Stirred Yogurt Treated with different Stabilizers for Improving Its Properties and Quality. Journal of Food and Dairy Science, 10 (12):473-478.

Hassanein, A. M., Yousef, E. T. and El-Shazly, H. A. (2014): Effect of concentrated pomegranate on probiotic yogurt. World Applied Science Journal, 30 (5): 567-574.

Hernández-Herrero, J. A. and Frutos, M. J. (2014): Effect of concentrated plum juice on physicochemical and sensory properties of yogurt made at bench top scale. International Journal of Dairy Technology, 67 (1): 123-128.

Hill, C., Guarner F., Reid, G., Gibson, R., Merenstein, D. J., Pot, B., Morelli, L., Canani, R. B, Flint, H. J., Salminen, S., Calder, P. C. and Sanders, M. E. (2014): The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. Nature Reviews Gastroenterology and Hepatology, 11 (8): 506-514.

International Dairy Federation (IDF) (1988): Yogurt: Enumeration of characteristics organism-colony count techniques at 37oC. Standard No.117A. Brussels, Belgium. International Dairy Federation, 117 (1): 1-4.

Ismail, E. A., Shenana, M. E., El-Alfy, M. B., Essawy, E. A. and Abd El-Halim, S. E. (2020): Novel probiotic adjunct cultures for the production of fruit flavored drinkable yogurt. Egyptian Journal of Food Science, 48 (2): 213-228.

Jaiswal, R., Karakose, H., Ruhmann, S., Goldner, K., Neumullerm M., Treutter, D. and Kuhnert, N. (2013): Identification of phenolic compounds in plum fruits (*Prunus salicina L.*) and *Prunus domestica L.)* by high performance liquid chromatography/tandem mass spectrometry and characterization of varieties by quantitative phenolic fingerprints. Journal Agriculture and Food Chemistry, 61 (49):12020-12031.

Jaroszewska, A. (2011): Quality of fruit cherry, peach and plum cultivated under different water and fertilization regimes. Journal of Elementolog, 16 (1): 51-58.

Jideani, A., Silungwe, H., Takalani, T., Omolola, A., Udeh, H. and Anyasi, T. (2021): Antioxidant-rich natural fruit and vegetable products and human health. International Journal of Food Properties, 24 (1): 41-67.

Johnson, J. B., Hoyos, B. E., Mani, J. S., Reynolds, M., Altvater, J. and Naiker, M. (2022): Identification of phenolics responsible for the high antioxidant activity in Burdekin plum (*Pleiogynium timoriense*) fruit. Food Chemistry Advances, 1 (1): 81-86.

Johnson, J. B., Collins, T., Mani, J. S. and Naiker, M. (2021): Nutritional quality and bioactive constituents of six Australian plum varieties. International Journal of Fruit Science, 21(1): 115-132.

Joseph, J. P. (2018): Probiotic genomes: Sequencing and annotation in the past decade. International journal of pharmaceutical sciences and research, 9 (4): 1351-1362**.**

Kasimoğlu, A., Goncuoğlu, M. and Akgun, S. (2004): Probiotic white cheese with *Lactobacillus acidophilus*. International Dairy Journal, 14 (12): 1067 -1073.

Kumar, A. and Kumar, D. (2016): Development of antioxidant rich fruit supplemented probiotic yogurts using free and microencapsulated *Lactobacillus rhamnosus* culture. Journal of Food Science and Technology, 53 (1): 667-675.

Lawrence, A. J. (1959): Syneresis of rennet curd. Part 1-Effect of time and temperature. Australian Journal of Dairy Technology, 14(4): 166-169.

Liguori, I., Russo, G., Curcio, F., Bulli, G., Aran, L., Della-Morte, D., Gargiulo, G., Testa, G., Cacciatore, F., Bonaduce, D. and Abete, P. (2018): Oxidative stress, aging, and diseases. Clinical Interventions in Aging, 13 (16): 757–772.

Ma, Y., Zhang, W., Cheng, S., Yang, W., Liu, Y., Yang, S., Zhang, X., Guo, M. and Chen, G. (2022): 1-methylcyclopropene treatment improves postharvest quality and antioxidant activity of *Prunus domestica L*. cv. Ximei fruit. Horticulture, Environment and Biotechnology, 63 (10): 857–867.

Mahmoud, A. E., Abebe, F. A., El-Demerdash, G. O. and Ekkehard, A. S. (2017): Antioxidant, anticancer and immunocytochemistry activities of three synthetic coumarin derivatives. Fayoum Journal of Agricultural Research and Development, 31 (2): 79-94.

Mannucci, C., Casciaro, M., Sorbara, E., Calapai, F., Di Salvo, E., Pioggia, G., Navarra, M., Calapai, G. and Gangemi, S. (2021): Nutraceuticals against oxidative stress in autoimmune disorders. Antioxidants, 10 (261): 1-28.

Marshall, R.T. (1992): Microbiological count methods. In: Standard methods for the examination of dairy products Marshall 16th ed. T.R., American Public Health Association, Washington, DC., USA.

Meenakshi, V. R., Ganya, S. and Umamaheswari, T. S. (2018): Formulation of value enriched probiotic fruit yogurt. International Journal of Current Microbiology and Applied Sciences, 7(3): 1440-1450.

Moludi, J., Alizadeh, M., Mohammadzad, M., and Davari, M. (2019): The effect of probiotic supplementation on depressive symptoms and quality of life in patients after myocardial infarction: Results of a preliminary double-blind clinical trial. Psychosomatic Medicine, 81 (9): 770-777.

Mradu, G., Saumyakanti, S., Sohini, M. and Arup, M. (2012): HPLC profiles of standard phenolic compounds present in medicinal plants. International Journal of Pharmacognosy and Phytochemical Research, 4 (3): 162-167.

Nasef, H. Y., Ibrahim, G. A., Shehata, A. N., Mohamed, A. F. and Farrag, A. A. (2020): Study of functional properties of probiotic and prebiotic on quality Characteristics of mango juice. Current Science International, 9 (3): 387-406.

Nyanzi, R., Elna, P. J. and Buys, E. M. (2021): Invited review: Probiotic yogurt quality criteria, regulatory framework, clinical evidence, and analytical aspects. Journal of Diary Science, 104 (1): 1-19.

Ordonez, A. A., Gomez, J. D., Vattuone, M. A. and Isla, M. I. (2006): Antioxidant activities of Sechium edule (Jacq.) Swart extracts. Food Chemistry, 97 (3): 452-458.

Pagthinathan, M., Nafees, M. S. and Powsika, E. (2018): Evaluate the physical, chemical and sensory parameters of probiotic yogurt during storage. AGRIEAST: Journal of Agricultural Sciences, 12 (2):10-18.

Perez-Fons, L., Garzon, M. T. and Micol, V. (2010): Relationship between the antioxidant capacity and effect of rosemary (*Rosmarinus officinalis* L.) polyphenols on membrane phospholipid order. Journal of Agriculture and Food Chemistry, 58 (1): 161-171.

Phaniendra, A., Jestadi, D. and Periyasamy, L. (2015): Free radicals: properties, sources, targets, and their implication in various diseases. Indian Journal of Clinical Biochemistry, 30 (1): 11-26.

Queen, B. L. and Tollefsbol, T. O. (2010): Polyphenols and aging. Current Aging Science, 3 (1): 34-42.

Qureshi, T. M., Nadeem, M., Ahmad, M. M., Hussain, S., Rehman, S. and Shaukat, A. (2017): Antioxidant potential of natural fruit flavored yogurt- a review. Journal of Agricultural Research, 55(1):85-99**.**

Rahman, M. T., Abu Zubair, M. D., Kamrunnahar, S. and Chakma, M. P (2020): Development of quality parameters for yogurt with strawberry juice. Food and Nutrition Science, 11 (12): 1070-1077.

Saleh, I., Abdelwahed, E. M., Rabie, A. M. H. and Abou El-Ella, W.M. (2018): Fortification of probiotic stirred yogurt by addition of apple and mango pulps. Food Dairy Home Economic Research, 45 (2):625-635.

Sallm, S. S., Hazaa, M. M., Eissa, H. A. A. , Metwally, M. R. and Ahmed, A. H. (2010): Evaluation of the microbiological quality of yoghurt blened with some fruit juice. Journal Food Dairy Sciences, Mansoura University, 1 (6): 337-353.

Sarvari, F., Mortazavian, A. M. and Fazeli, M. (2014): Biochemical characteristics and viability of probiotic and yogurt bacteria in yogurt during the fermentation and refrigerated storage. Applied Food Biotechnology, 1 (1): 55-61.

Shahbandari, J., Golkar, A., Taghavi, S. M. and Amiri, A. (2016): Effect of storage period on physicochemical, textural, microbial and sensory characteristics of stirred soy yogurt. International Journal of Farming and Allied Sciences, 5 (6): 476-484.

Sharifi-Rad, M., Kumar, N. V., Zucca, P., Varoni, E. M., Dini, L., Panzarini, E., Rajkovic, J., Fokou, P. V., Azzini, E., Peluso, I, Mishra, A. P., Nigam, M., El Rayess, Y., El Beyrouthy, M., Polito, L., Iriti, M., Martins, N., Martorell, M., Docea, A. O., Setzer, W. N., Calina, D., Cho, W. C. and Sharifi-Rad, J. (2020): Lifestyle, Oxidative Stress, and Antioxidants: Back and forth in the pathophysiology of chronic diseases. Frontiers in Physiology, 11 (1): 694-715.

Silva, F. A., De Oliveira, M. E., de Figueiredo, R. M., Sampaio, K. P., de Souza, E. L., de Oliveira, C. E., Pintado, M. M. and Queiroga, R. R. (2017): The effect of Isabel grape addition on the physicochemical, microbiological and sensory characteristics of probiotic goat milk yogurt. Journal of Food and Function, 8 (6): 2121-2132.

Singleton, V. L. and Rossi, J. A. (1965): Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American Journal of Enology and Viticulture, 16 (3): 144-158.

Taneva, I. and Zlatev, Z. (2020): Total phenolic content and antioxidant activity of yogurt with Goji berries (lycium barbarum): Scientific Study and Research: Chemistry and Chemical Engineering. Journal of Food Industry, 21 (1):125-131.

Terzaghi, B. E. and Sandine, W. E. (1975): Improved medium for lactic streptococci and their bacteriophages. Journal of Applied Microbiology, 29 (6): 807-813.

Tripathi, M. K and Giri, K. (2014): Probiotic functional foods: Survival of probiotics during processing and storage. Journal of Functional Foods, 9 (1): 225-241.

Vital, A. C., Goto, P. A., Hanai, L. N., Gomes-da-Costa, S. M., Filho, B. A., Nakamura, C. V. and Matumoto-Pintro, P. T. (2015): Microbiological, functional and rheological properties of low fat yogurt supplemented with *Pleurotus ostreatus* aqueous extract Lebensmittel-Wissenschaft und Technologie - Food Science and Technology, 64 (2): 1028-1035.

Vitheejongjaroen, B., Kanthawang, P., Loison, F., Jaisin, F., Pachekrepapol, U. and Taweechotipatr, M. (2021): Antioxidant activity of Bifidobacterium animalis MSMC83 and its application in set-style probiotic yogurt. Food Bioscience, 43 (1): 101259-101267.

Wang, Y., Wu, Y., Wang, Y., Xu, H., Mei, X., Yu, D., Wang, Y. and Li, W. (2017): Antioxidant properties of probiotic bacteria. Nutrients, 9 (5): 1-15.

Widyastuti, Y., Febrisiantosa, A. and Tidona, F. (2021): Health - Promoting Properties of Lactobacilli in Fermented Dairy Products. Frontiers in Microbiology, 12 (6): 673890-673897.

Yagmur, C. and Taskin, M. (2011): Study on changes in mineral content of plum (*Prunus domestica*) and strawberry (Fragaria × ananassa) during canning. Indian Journal of Agricultural Sciences, 81 (8): 723-728.

Zhou, R., Liu, X., Chen, L., Huang, J., Liang, X., Wan, D., Zhang, S. and Huang, L. (2020): Comparison of the antioxidant activities and phenolic content of five Lonicera flowers by HPLC-DAD/MS-DPPH and chemometrics. International Journal of Analytical Chemistry, 20 (2): 111-116.

تأثير البروبيوتك على النشاط المضاد للأكسدة للمركبات الفينولية في الزبادي الوظيفي المدعم بعصير البرقوق

المستخلص

تعتبر منتجات الألبان مناسبة للوجبات الوظيفية نظرا لقيمتها الغذائية العالية ويتميز الزبادي بقيمته الغذائية المرتفعة عن باقي منتجات الألبان المتخمرة الأخرى وتساعد منتجات الألبان المتخمرة في المحافظة على صحة الجهاز الهضمي. هدفت هذه الدراسة إلى بحث تدعيم الزبادي المصنوع من لبن البقر ب ١٠ % عصير برقوق وإضافة بكتريا *bifidobacterium* كبكتريا بروبايوتيك مع بكتريا البادئ العادية *Lactobacillus delbrueckii subsp. Bulgaricus* و *Streptococcus salivarius subsp. Thermophilus.* تم تقدير كلا من الاختبارات الفيزيوكيميائية، الريولوجية، البيوكيميائية والعد الميكروبي خلل فترات التخزين ١، ٧، ١٤ يوم على درجة حرارة ٤ درجة مئوية. ويتضح من النتائج أن زبادي البروبايوتيك المدعم بعصير البرقوق كان له اعلى معدل حموضة ومواد صلبة كلية بينما كان الأقل في معدل الأس الهيدروجيني، الدهون، البروتين والرماد بالمقارنة بالزبادي الكنترول. النسب المئوية لقيم التركيب الكيميائي لكلا من المواد الصلبة الكلية، الدهون، البروتين والرماد انخفضت خلال فترات التخزين. إضافة عصير البرقوق بتركيز ١٠٪ سبب حدوث أعلى كمية للشرش لكل المعاملات، والتي تراوحت من ١٦,٣ مل في اليوم ١ حتى ٢٢,٢ مل في اليوم ال ١٤ وأقل معدل لزوجة لكل المعاملات، والتي تراوحت من ١٤٩±١٤سنتيبويز في اليوم ١ حتى ٧٠±٩ سنتيبويز في اليوم ال ١٤و الذي بدوره أثر على القبول العام للزبادي والذي يتأثر أيضا بمدة التحزين. وبينت النتائج أيضا أن ellagic acid كان أكثر المركبات الفينولية الشائعة الموجودة في عصير البرقوق (٢٨,٣٧±٠,٠١ ميكروجرام/مل) ، بينما gallic acid في الزبادي الكنترول، و chlorogenic acid في زبادي البروبايوتيك و quercetin في الزبادي المدعم بعصير البرقوق و catechin في زبادي البروبايوتيك المدعم بعصير البرقوق. بالرغم من ان إضافة عصير البرقوق للزبادي قد عزز من محتوى الفينولات الكلية ومحتوى الفلافونيدات الكلية وكذلك النشاط المضاد للأكسدة ولكنه تسبب في خفض خصائص التماسك والقوام. وأكثر من ذلك، فإن تدعيم الزبادي بعصير البرقوق بإضافة *Bifidobacterium* أو بدونها كان يحتوي على أقل عدد للبكتريا مقارنة بالزبادي بدون عصير البرقوق.

الاستنتاج: نتائج هذه الدراسة أشارت إلى أن تدعيم الزبادي بعصير البرقوق مع *Bifidobacterium* وبكتريا البادئ العادية له العديد من الفوائد الصحية وخصوصا نشاطه

المضاد للأكسدة والمركبات الفينولية والتى لها تأثير وقائي مضاد للعديد من الأمراض كما ومن جهة آخرى فهو يحسن من جودة الزبادي ويعطي بدائل متنوعة عديدة للمستهلكين.

الكلمات المفتاحية: زبادي، عصير برقوق، المركبات الفينولية، النشاط المضاد للأكسدة، بكتريا البروبايوتيك، التقييم الحسي، اللزوجة والشرش.