



Nutritional, antioxidant, and antimicrobial assessment of carrot powder and its application as a functional ingredient in probiotic soft cheese

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ABSTRACT

Carrots (the main source of carotenoids) have multiple nutritional and health benefits. The objectives of this study were to evaluate the compositional, antioxidant, and antimicrobial properties of carrot powder and to examine its effect on the sensory characteristics, chemical properties, and microbial viability of probiotic soft cheese at a rate of 0.2, 0.4, and 0.6%. The carrot was turned into powder before being analyzed and incorporated as an ingredient in making probiotic soft cheese. Probiotic soft cheese was made from buffalo milk. The buffalo milk (~6.9% fat, 4.4% protein, 9.2% milk solids not fat, and 0.7% ash) was pasteurized at $75 \pm 1^\circ\text{C}$ for 5 min and cooled to $40\text{--}42^\circ\text{C}$. The milk was then divided into 4 aliquots. Sodium chloride (local market, Assiut, Egypt) was added at a ratio of 5% followed by starter cultures. The carrot powder (4.5% moisture, 4.8% ash, 2.7% fat, 8.2% protein, 11.9% fibers, and 72.3% carbohydrate) was added at a rate of 0.2, 0.4, and 0.6%, followed by addition of 0.02 g/kg rennet. The cheese was cut again into cubes, pickled in jars filled with whey, and stored for 28 d at $6 \pm 1^\circ\text{C}$. The results of this study illustrated the nutritional and antioxidant properties of carrot powder. Incorporation of carrot powder in probiotic soft cheese affected the moisture and salt content at 0 d. The total bacteria count decreased from 7.5 to 7.3 log cfu/g in the cheese when carrot powder was used at a rate of 0.6%. The reduction of total bacteria count was noticed during the 28 d of storage by adding carrot powder. Furthermore, lactic acid bacteria and *Bifidobacterium longum* counts elevated with adding carrot powder during the 28 d of storage.

Key words: carrot powder, nutritional, antioxidant, antimicrobial, probiotic soft cheese

INTRODUCTION

Due to the high nutritional value of food items with added functional ingredients, the demand for those types of foods has increased significantly. The presence of food containing phytochemicals in our daily diet may reduce birth defects, cancer, cardiovascular, and neurodegenerative diseases (Blancquaert et al., 2010; Liu, 2013). Carrot, being a good source of phytochemicals (e.g., carotenoids, phenolics, ascorbic acid, and polyacetylenes; Ahmad et al., 2019), is associated with reducing the risk of cardiovascular disease and cancer.

Carrot (*Daucus carota* L.) is a nutritious root vegetable (seasonal crop), which is not available throughout the year. Drying the carrot could be an efficient way to extend its shelf-life (Al-Amin et al., 2015). Carrots are known as a multnutritional food source and are rich in natural bioactive compounds, such as phenolics, carotenoids, polyacetylenes, and ascorbic acid (Ahmad et al., 2019), fiber, and minerals. As a result, carrots can be used as a functional ingredient in any product to increase the biological and nutritional values (Ergun, 2018). Moreover, several studies used carrot juice to develop probiotic beverages (Duda-Chodak et al., 2015; El-Sayed et al., 2016; Rafiq and Sharma, 2016) to be used as suitable media for the cultivation of probiotics. Strains of *Bifidobacterium* and *Lactobacillus* genera are commonly considered under Generally Recognized As Safe status and are used as functional probiotic dairy products (De Souza Oliveira et al., 2009; Awasti et al., 2016). Furthermore, many studies used probiotic bacteria in cheese making due to their role in enhancing flavor, texture, and shelf-life (Songisepp et al., 2004; Hammam and Ahmed, 2019; Ahmed et al., 2021; Hamdy et al., 2020), while the addition of natural bioactive components in cheese could improve

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the nutritional, functional, and sensory characteristics (Ritota and Manzi, 2020). Likewise, using tomato powder as an ingredient in processed cheese formulations resulted in better functional properties in the final product with higher antioxidant activity, as well as sensory properties (Solhi et al., 2020). Another study found that addition of aqueous extracts of the *Inula britannica* flower in making cheese elevated the protein, ash, and total phenolic content of final cheese (Lee et al., 2016). Furthermore, utilization of basil leaves in Serra da Estrela cheese led to higher antioxidant activity and kept unsaturated fatty acid and protein contents far from deuteriation (Carocho et al., 2016). Moreover, adding celery leaf during the making of soft white cheese contributed to better flavor and overall acceptability (Awda et al., 2019). In contrast, others found that adding 0.3% (wt/vol) of turmeric powder decreased the score of flavors (Al-Obaidi, 2019).

In recent years, several studies emphasized the health benefits of probiotic cheese. To date, no study used carrot powder to fortify probiotic soft cheese. Therefore, this work aimed to evaluate the nutritional and antioxidant properties of carrot powder and to examine the effect of the carrot powder on the compositional and functional characteristics of soft cheese during storage for 28 d.

MATERIALS AND METHODS

No human or animal subjects were used, so this analysis did not require approval by an Institutional Animal Care and Use Committee or Institutional Review Board.

Preparation of Carrot Powder

Carrots were obtained from a local market (Assiut, Egypt) and washed thoroughly with water and cut into thin slices. Those slices were steam blanched for 3 min to inactivate enzymes (pectinase and peroxidase) and elevate the bioavailability of beta-carotene by thermal treatment (Ghosh et al., 2019). The slices were then dried in an electric oven at 55°C for 12 h. The dried carrot was then turned into powder using a high speed grinder (DeLonghi). Subsequently, the carrot powder was sterilized at 121°C for 11 min.

Cheese Manufacturing

Lactococcus lactis ssp. *lactis* ATCC (11454), *Lactiplantibacillus plantarum* ssp. *plantarum* (ATCC 14917), *Lactococcus lactis* ssp. *cremoris* (ATCC 19257), and *Bifidobacterium longum* (ATCC 15707)

were obtained from the Egyptian Microbial Culture Collections (Cairo Microbiological Resources Center, Faculty of Agriculture, Ain Shams University, Cairo, Egypt). *Lactiplantibacillus plantarum* ssp. *plantarum* was activated using anaerobic conditions in sterile skim milk at 37°C for 24 h., while *Lactococcus lactis* ssp. *lactis* and *Lactococcus lactis* ssp. *cremoris* were activated at 30°C for 24 h. The viable count of the mix of *Lactococcus lactis* ssp. *lactis* and *Lactococcus lactis* ssp. *cremoris* was 7.5 ± 0.09 log cfu/mL, whereas *Lactiplantibacillus plantarum* ssp. *plantarum* was 7.5 ± 0.02 log cfu/mL, and *Bifidobacterium longum* was 7.6 ± 0.28 log cfu/mL.

Probiotic soft cheese was made from buffalo milk as described in a previous study (Mohammed et al., 2016a) with some modifications. Fresh buffalo milk (6.93% fat, 4.37% protein, 9.23% milk SNF, and 0.68% ash) was obtained from the Animal Production Farm (Faculty of Agriculture, Assiut University, Assiut, Egypt). The buffalo milk was heated to $75 \pm 1^\circ\text{C}$ for 5 min and then cooled to 40–42°C. The milk was then divided into 4 aliquots. Sodium chloride (local market, Assiut, Egypt) was added to each portion at a ratio of 5%, and then the mix of *Lactococcus lactis* ssp. *lactis* and *Lactococcus lactis* ssp. *cremoris* was added at 2%, followed by the addition of 1% *Lactiplantibacillus plantarum* ssp. *plantarum*, and 15% *Bifidobacterium longum*. The inoculated milk was left for 30 min. The first portion was used as a control, whereas the other 3 portions were supplemented with 0.2, 0.4, and 0.6% carrot powder. Subsequently, rennet (Chr. Hansen's) was added at a rate of 0.02 g/kg. The fermentation was done at 40–42°C until the curd was set. The cheese was then cut and left in cheesecloth to drain for 3 d at 5°C. The cheese was cut again into cubes, pickled in plastic jars filled with whey, and stored for 28 d at $6 \pm 1^\circ\text{C}$. This experiment was repeated 3 times. The yield of cheese samples was calculated as the weight of cheese divided by the weight of milk expressed as a percentage (Mehaia, 1993).

Compositional Analyses

Most of the chemicals were obtained from El-Gomhouria Inc. The 2,2-diphenyl-1-picrylhydrazyl (DPPH), Folin-Ciocalteu reagent, and gallic acid were procured from Sigma-Aldrich.

Chemical Composition

Moisture, fat, and total protein (TP) were determined (AOAC International, 2000) in carrot powder and cheese samples. Ash, crude fiber (AOAC Interna-

tional, 2000), carbohydrates (Pellet and Shadarevian, 1970), and caloric value (Livesey, 1995) were estimated in carrot powder. Titratable acidity (AOAC International, 2000), salt (Bradley et al., 1992), soluble nitrogen (SN), and NPN were determined in cheese samples using the Kjeldahl method (AOAC International, 2000). Those analyses were duplicated on each replicate.

Determination of Mineral Profile

The calcium, magnesium, iron, and zinc were determined by iCAP6000 (Thermo Scientific) inductively coupled plasma emission spectrometry (Isaac and Johnson, 1985). Sodium and potassium contents were determined using a Flame Photometer Corning 400 (Jenway; Chapman and Pratt, 1962), while phosphorus was measured by a spectrophotometer (Jackson, 1967). The mineral profile was performed on 2 samples of each replicate.

Determination of Vitamin C

Vitamin C (ascorbic acid) content was determined by titration using the iodine solution method (Satpathy et al., 2020). Vitamin C was measured in duplicates on each replicate.

Determination of Total Carotenoid by Spectrophotometer

Extraction and determination of total carotenoid were measured (Al-Dabbas et al., 2015), while vitamin A was measured as the retinol equivalent, where 1 retinol equivalent = 1 µg of retinol = 6 µg of β-carotene (Pitchiah, 2004). Total carotenoid was measured 2 times on each replicate.

Determination of Total Phenolic Compounds

Total phenolics content was measured (Singleton et al., 1999) and the results were expressed as milligrams of gallic acid equivalents per 100 g sample (mg of GAE/100 g sample). This test was duplicated on each replicate.

Antioxidant Activity

The antioxidant activity of carrot powder extract was estimated using DPPH scavenging activity (Po-thitirat et al., 2009). This was done in duplicates. As DPPH solution in methanol (0.13 mM) was added to the methanolic extract of carrot powder, the control sample was prepared by mixing methanol and DPPH.

Then mixtures were allowed to stand in the dark at room temperature for 30 min. The absorbance of the mixture was recorded at 517 nm by spectrophotometer, and the ability of carrot powder extract to scavenge DPPH radical was calculated as follows:

$$\text{DPPH} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100.$$

Gas Chromatography-Mass Spectrometry Analysis

The GC-MS analysis was conducted to analyze volatile and semi-volatile compounds (Department of Analytical Chemistry, Faculty of Science, Assiut University, Assiut, Egypt). The extract was prepared in chloroform as follows: 1 g of carrot powder, 0.1 mL of phosphoric acid (1:1), and 2 mL of chloroform were added. Then it was sonicated for 15 min by ultrasonic, followed by centrifugation ($5,009 \times g$) for 20 min at 15°C. Subsequently, the clear organic layer was separated and injected into GC-MS. The GC-MS analysis was performed using a GC-MS (7890A-5975B) equipped with column DB 5 ms (30 m $\times$ 0.250 mm $\times$ 0.250 µm). The initial temperature of the GC oven was kept at 60°C and then increased to 120°C at a rate of 10°C/min, followed by a series of continuous temperature increase to 175°C at a rate of 3°C/min, 205°C at a rate of 5°C/min, 210°C at a rate of 0.8°C/min, and finally at 280°C at a rate of 5°C/min. Helium was used as the carrier gas at a flow rate of 0.5 mL/min. Volatiles were identified using the Wiley registry, 11th ed, and the NIST 2017 mass spectral library. Data are expressed as the percentage of the total GC area. Duplicate samples were tested for each replicate.

Microbiological Analysis

Antimicrobial Activity. The antimicrobial activity of the carrot powder was tested against 6 bacterial strains (*Bacillus cereus* AUMC no. B-52, *Escherichia coli* AUMC no. B-53, *Micrococcus luteus* AUMC no. B-112, *Pseudomonas aeruginosa* AUMC no. B-73, *Serratia marcescens* AUMC no. B-55, and *Staphylococcus aureus* AUMC no. B-54) obtained from Mycological Center, Faculty of Science, Assiut University, Assiut, Egypt. Bacterial strains were individually inoculated in a nutrient broth medium for 48 h to prepare inoculum. Sterile plastic Petri plates (10 cm diameter) and nutrient agar medium were used in the bioassay (Johnson and Case, 2018). After media solidification, a sterile cork borer was used to cut 5-mm diameter cavities in the solidified agar (4 cavities per plate). Carrot powder

Table 1. Mean (n = 3) composition of carrot powder used in making probiotic soft cheese

Sample	Chemical composition ¹ (g/100 g on a dry basis)						Mineral (mg/100 g on a dry basis)							Caloric value ² (kcal/100 g)
	Moisture	Ash	Fat	TP	Fiber	Carbs	Ca	Na	K	P	Mg	Fe	Zn	
Carrot powder	4.55	4.80	2.75	8.25	11.94	72.26	880.0	615.0	532.7	199.3	171.7	20.7	3.4	346.76
SD	0.01	0.02	0.03	0.01	0.03	0.05	3.0	4.0	0.2	0.2	0.2	0.1	0.3	0.11

¹TP = total protein; carbs = carbohydrates.²Caloric value = (TP × 4) + (carbs × 4) + (fat × 9).

methanolic extract was pipetted in the cavities (20 µL per cavity). Plates were incubated at 28°C for 48 h, and the diameter of the inhibition zone around cavities was measured and reported in mm (Kwon-Chung and Bennett, 1992). This was done on 2 samples of each replicate.

Microbiological Count. Bacterial counts were performed in cheese samples at 0 and 7, 14, 21, and 28 d. Total bacteria count (**TBC**) was enumerated on a nutrient agar medium (Plate Count Agar, HiMedia Laboratories Pvt. Ltd.) after incubation at 37°C for 48 h (Suleiman et al., 2011) using the standard plate count technique (Wehr and Frank, 2004). Lactic acid bacteria (**LAB**) counts were enumerated on a de Man, Rogosa, and Sharpe agar medium (Neogen culture media) after incubation anaerobically at 37°C for 48 h. *Bifidobacterium* counts were enumerated on a modified de Man, Rogosa, and Sharpe medium (Renschler et al., 2020) after incubating plates anaerobically at 40°C for 48 h. Skim milk agar medium was used in the enumeration of proteolytic bacteria (Awad et al., 2005) after incubation at 37°C for 4 d. Furthermore, psychrophilic bacteria (Baumann and Reinbold, 1963) and lipolytic bacteria (Davis and Ewing, 1964) were enumerated. Molds were enumerated using Czapek's dextrose agar medium (Pitt and Hocking, 2009), while malt extract agar medium (Yousif and Safaa, 2014) was used for yeast enumeration. Two samples were analyzed for microbiological analyses in each replicate at each time point.

Sensory Analyses. Cheese samples were judged by 10 staff members of the Dairy Science Department at Assiut University for sensory analyses. Flavor (50 points), body and texture (40 points), and color and appearance (10 points) were evaluated at 0 and 28 d of storage to have 100 points total (Bodyfelt and Potter, 2008).

Statistical Analyses. One-way (treatment) and 2-way (treatment and time) ANOVA were tested using SPSS software (version 16.0 for Windows, SPSS Inc.). When significant differences ($P < 0.05$) were noticed among means, Duncan's multiple comparison test was used for mean difference testing.

RESULTS AND DISCUSSION

Composition of Carrot Powder

The chemical composition of carrot powder samples is shown in Table 1. The moisture content of carrot powder was 4.5%, whereas ash, fat, TP, and fiber contents were 4.80, 2.75, 8.25, and 11.94% on a dry basis, respectively. It was found that ash, fat, TP, and fiber in carrot powder were approximately 6.80, 2.90, 9.20, and 7.20%, respectively, on a dry basis (Hussein et al., 2013). Another study stated that carrot samples have moisture content of 11.70%, whereas ash, fat, TP, and fiber contents were approximately 13.40, 5.40, 10.40, and 13.70% on a dry basis, respectively (Mandey et al., 2019). The total carbohydrate content of carrot powder in current research recorded was around 72.30%, while the caloric value was 346.8 kcal/100 g on a dry basis. Our results were in agreement with those reported by Singh et al. (2013), as they observed that carbohydrate and caloric values in carrot powder were 68.3% and 336.0 kcal/100 g on a dry basis, respectively.

Potassium, phosphorus, calcium, sodium, magnesium, iron, and zinc contents in carrot powder are illustrated in Table 1. In the present study, potassium, phosphorus, calcium, sodium, magnesium, iron, and zinc were approximately 532.7, 199.3, 880.0, 615.0, 171.7, 20.7, and 3.4 mg/100 g on a dry basis, respectively. Similar findings were exemplified by Singh et al. (2001), as they found that zinc content in carrots was 3.2 mg/100 g on a dry basis. Moreover, it was reported that calcium, phosphorus, and iron contents in carrot pulp waste were 314.95, 317.77, and 12.65 mg/100 g on a dry basis, respectively (Shyamala and Jamuna, 2010).

Phytochemical Components in Carrot Powder

Vitamin C, β-carotene, vitamin A, total phenolics, and antioxidant activity of carrot powder samples are presented in Table 2. Data showed that vitamin C content was 50.0 mg/100 g on a dry basis. It was reported that ascorbic acid was 8.0 mg/100 g of fresh orange-colored carrots (Thamburaj and Singh, 2001).

Table 2. Phytochemical components (on a dry basis) of carrot root powder¹

Sample	Vitamin C (mg/100 g)	β -carotene (mg/100 g)	Vitamin A (RE)	Total phenolics (mg of GAE/100 g)	Antioxidant activity (%)
Carrot powder	50.0	123.0	20,500.0	203.6	64.45
SD	4.0	5.0	4.0	0.2	0.05

¹RE = retinol equivalent; GAE = gallic acid equivalents.

Furthermore, in the current study, β -carotene content was 123.0 mg/100 g on a dry basis. However, another study observed lower content of β -carotene in carrot powder at a rate of 1,154.20 μ g/g or 115.42 mg/100 g (Pitchiah, 2004), whereas another reported higher β -carotene (254.0 mg/100 g) on a dry basis (Béttega et al., 2014) when compared with our findings. Such differences could be attributed to the variety and environmental factors. Furthermore, The retinol equivalent of carrot powder (Table 2) was about 20,500 (RE/100 g), which was obtained using the standard conversion formula. Likewise, others studies examined 5 groups of fresh carrots and found that the β -carotene and vitamin A contents were 12,300 μ g/100 g and 2,054.1 RE/100 g, respectively (Aremu and Nweze, 2017).

The total phenolics content (Table 2) in carrot powder was 203.6 mg/100 g on a dry basis sample, whereas it was reported that the range of total phenolic content in 15 varieties of orange carrot ranged from 18.7 to 33.8 mg/100 g as gallic acid equivalents on a fresh weight basis (Leja et al., 2013).

The antioxidant activity of carrot powder (determined by DPPH scavenging activity) was 64.45%. Our value is higher than the values reported in the study conducted by John et al. (2017), which ranged from 7.45 to 34.9%. Another study by Leja et al. (2013) estimated radical scavenging activity using a stable DPPH radical. They found that the DPPH ranged from 3.5 to 13.7% in 15 varieties of orange carrot extracts (on a fresh weight basis). Moreover, the importance of carotenoids (which are located in orange carrots) was referred to as a valuable antioxidant component that can neutralize the effect of free radicals (da Silva Dias, 2014).

Gas Chromatography-Mass Spectrometry Analysis

The GC-MS of compounds was carried out in chloroform extract of carrot powder. The GC-MS in our study detected 43 compounds as shown in Table 3; however, a previous study detected 18 compounds using the same methodology (Gaggiotti et al., 2019). The retention time and value (% concentration) in Table 3 indicated that the first setup peak was tetrahydro-2-(3-methyltricyclo[2.2.1.0(2,6)] hept-3-yl)oxyl-2H-pyran. The most volatile compounds were butyl-9,12-octadecadie-

noate; glycerol 1-palmitate, O-cyanobenzaldehyde; and 7.7',8,8',11,11',12,12',15,15'-decahydro-beta,psi-carotene. Moreover, the data in Table 3 detected terpenoids in carrot powder samples, which were alpha-terpinene and (E)-geranylacetone (monoterpenes); caryophyllene, *trans*-gamma-bisabolene and 3 β -hydroxy-8-oxo-6. β .H.7. α .H, 11. β . H-germacran-4(14),9(10)-dien (sesquiterpene); phytane (diterpenes); and squalene (triterpenes). Our data were similar to previous studies, which concluded that the terpenes were the most important volatiles identified in carrots (especially in red and orange carrots; Güler et al., 2015; Gaggiotti et al., 2019). A different study examined the volatile compounds in carrot juice from 12 different varieties and found alpha-terpinene (2.51, 2.69), geranylacetone (2.06, 2.69), caryophyllene (3.94, 3.53), and (E)-gamma-bisabolene (4.05, 4.34) in fresh and processed carrots, respectively (Fukuda et al., 2013). Table 3 reveals that *trans*-beta-ionone recorded 3.78%, which is considered as a transconfiguration of β -ionone (cyclic terpenoid required for vitamin A synthesis; Fahlbusch et al., 2003), whereas *cis*-p-mentha-2,8-dien-1-ol (monocyclic terpene) found at a concentration of 0.19%. Additionally, many terpenes are known to have antimicrobial, anticarcinogenic, and antidepressant activities, and they are associated with reducing plasma glucose levels in diabetes mellitus rats (Himejima et al., 1992; Myers et al., 1997; Gupta et al., 2007; Bahramsoltani et al., 2015). It was reported that geranyl acetone has high antiradical activity (Stobiecka, 2015). Moreover, squalene is considered a precursor of steroids, and it stimulates the decrease of total cholesterol, LDL cholesterol, and triacylglycerol levels in hypercholesterolemic patients (Miettinen and Vanhanen, 1994; Chan et al., 1996). In addition, squalene participates in the synthesis of vitamin D in the human body (Campos-Vega and Oomah, 2013). However, Table 3 shows that 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-2(4H) benzofuranone and diethylmalonic acid monochloride tetrahydro furfuryl ester recorded 1.68 and 0.95%, respectively, which were related to thermal processing in dehydrated carrots (Soria et al., 2008). Ethyl-8H-naphtho[2', 1:5,6] pyrano[4,3-b] quinoline-11-carboxylate and 6,8-dichloro-2-[4-chlorophenyl]-4-bromoacetyl quinoline in Table 3 recorded 1.78 and 1.01%, respectively, and belong to a group of quinoline derivatives, which are consid-

ered important fragrance substances (Fahlbusch et al., 2003). They also have antioxidant, antitumor, and anti-malarial activities (Abas et al., 2006; Behforouz et al., 2007; Kaur et al., 2009). Also, Table 3 indicates several fatty acids and their esters in carrot powder samples, similar to 9,12-octadecadienoic acid (z,z)-,2(acetyloxy)-1-[(acetyloxy)methyl] ethyl ester, tetradecanoic acid, 1-methylethyl ester, hexadecanoic acid methyl ester, hexadecanoic acid, ethyl ester, *trans*-delta(sup 9)-octadecenoic acid, oleic acid, 9-octadecenoic acid (Z)-, eicosyl ester, octadecanoic acid, methyl ester, 3-methyl-2,6-dioxo-4-hexenoic acid, delta 9-*cis*-oleic acid, butyl 9,12-octadecadienoate and (6Z)-6-octadecenoic acid. While the compound 6-fluoro-7-dehydrocholesterol was detected (0.30%) in carrot powder samples (Table 3), it also plays an important role in the synthesizing of vitamin D (Miguel et al., 2019). Data in Table 3 revealed that faltarinol recorded 2.01%. Also, it was found that carrot extracts contain different amounts of faltarinol, faltarindiol, and faltarindiol 3-acetate (Purup et al., 2009). In addition, another study revealed that faltarinol had anticancer properties (Zidorn et al., 2005). Table 3 shows that eicosane content recorded 0.43%, which has very strong anti-inflammatory effects (Okechukwu, 2020). Also, it was shown that eicosane had an antimicrobial and antifungal effect (Ahsan et al., 2017). Estra-1,3,5(10)-trien-17 β -ol in Table 3 recorded 2.93%, which has antitumor, anti-inflammatory, antioxidant, and antimicrobial activities (Mohammed et al., 2016b). In contrast, Table 3 shows that carrot powder contains 1-(4-bromobutyl)-2-piperidinone, which has a strong effect on the pathogenic microorganisms (Al-Salman, 2019), whereas estra-1,3,5(10)-trien-17 β -ol in our research recorded 2.93%, which has antitumor, anti-inflammatory, antioxidant, and antimicrobial activities (Mohammed et al., 2016b). Moreover, 2-fluorophenyl methyl-1H-purin-6-amine in Table 3 recorded 3.69%, which has antioxidant properties (Budayatin et al., 2021). However, 1,2,3,4-tetrol-L-ribo-octadecane was 3.08%, belonging to guggultetrols, which has a potential treatment for type-II diabetes (Angadi et al., 2013).

Antimicrobial Activity

The antimicrobial potential of carrot powder extracts against 6 pathogens was examined in terms of zone inhibition of bacterial growth, and the results are exemplified in Table 4. The growth inhibition zone was measured for 50 μ L of carrot powder methanolic extracts, and the zones were in the range of 7.0–10.0 mm. The maximum zone formation was against *Pseudomonas aeruginosa*. According to a previous study (John et al., 2017), it was observed that the growth inhibition zone of the methanolic extracts of carrot peel for 4 bacte-

ria (*Shigella flexneri*, *Escherichia coli*, *Staphylococcus aureus*, and *Klebsiella pneumonia*) ranged from 12 to 14 mm. These differences in zone inhibition of carrot extracts could be attributed to the variation in sample extract concentrations.

Probiotic Soft Cheese: Cheese Yield

Figure 1 demonstrates the cheese yield of probiotic soft cheese. The cheese yield ranged from 18.2 to 20.5%. These results were lower than the yield of fresh soft cheeses made from cow's milk with different systems (monoculture system and intensive silvopastoral system in different seasons; Mohammed et al., 2016a). The presence of somatic cell proteinases in milk could be responsible for these differences (Holt and Roginski, 2001). According to Figure 1, the carrot addition in probiotic soft cheese samples reflected a significant ($P < 0.05$) increase in cheese yield. This increase in cheese yield could be associated with the presence of fiber content in carrot powder, containing hydroxyl groups that interact with the hydrogen bonds of water (Gómez et al., 2011). This, in turn, increases the water holding capacity (Guyih et al., 2020), and thereby elevates the moisture content and yield of the final cheese. Emmons (1993) found that increasing the moisture content in cheese from 0.32 to 0.57% elevated the yield from 1.67 to 2.87%. Mehaia (1993) reported that low cheese yield might be caused by lower moisture content in the cheese.

Properties of Probiotic Soft Cheese

Sensory Evaluation. Sensory evaluation results for fresh cheese and after 28 d of storage are shown in Table 5 and Figure 2. According to Table 5, probiotic cheese with 0.2% carrot powder recorded the highest values in all studied parameters after 28 d, but this increase was not significant ($P > 0.05$). Carrot addition enhances the flavor due to its content of terpenoids and sugars, which are responsible for carrot flavor (Jones, 2008). Although we observed an increase in the additional percentage of carrot powder during cheese making to 0.6%, body and texture parameters decreased which resulted in holes and increased firmness in the final cheese. In terms of color and appearance, T3 (probiotic cheese with 0.6% carrot powder) had a slight orange color. This observation of color was reported in another study (Hussein et al., 2013). With the increase in the storage duration and ripening of cheese samples, we observed an increase in the overall sensory characteristics of all cheese samples as illustrated in Table 5, which was similar to another report (Hamdy et al., 2020). They noticed enhancement of flavor, body, and texture of

Table 3. Gas chromatography-mass spectrometry analysis results of carrot powder methanolic extract

No.	Retention time (min)	Compound	Value (%)
1	13.703	Tetrahydro-2-[(3-methyltricyclo[2.2.1.0(2,6)] hept-3-yl)oxyl-2H-pyran	0.214
2	14.207	Caryophyllene	0.842
3	14.285	Alpha terpinene	1.630
4	14.66	(E)-geranylacetone	1.475
5	15.372	<i>Trans</i> -beta-ionone	3.781
6	15.695	O-cyanobenzaldehyde	4.116
7	16.083	<i>Trans</i> -gamma-bisabolene	0.682
8	17.138	5,6,7,7a-tetrahydro-4,4,7a-trimethyl-2(4H) benzofuranone	1.681
9	17.474	Diethylmalonic acid, monochloride, tetrahydrofurfuryl ester	0.955
10	17.875	Ethyl-8H-naphtho[2', 1:5,6]pyrano[4,3-b] quinoline-11-carboxylate	1.779
12	20.624	Phytane	0.241
13	21.084	9,12-octadecadienoic acid (z,z)-,2(acetyloxy)-1-[(acetyloxy)methyl] ethylester	0.474
14	22.015	6,8-dichloro-2-[4-chlorophenyl]-4-bromoacetyl quinoline	1.013
15	22.339	<i>Cis</i> -p-mentha-2,8-dien-1-ol	0.190
16	23.348	Tetradecanoic acid, 1-methylethyl ester	1.753
17	24.15	6-fluoro-7-dehydrocholesterol	0.298
18	24.318	Cystine, Tbs 2x	1.148
19	24.725	<i>Trans</i> -geranylgeraniol	1.867
20	24.874	Hexadecanoic acid methyl ester	2.097
21	25.01	Squalene	0.280
22	25.489	2-methyl-6-tert-octylphenol	0.657
23	25.612	3 beta-hydroxy-8-oxo-6,β.H.7.α.H, 11.β. H-germacran-4(14),9(10)-dien	0.431
24	25.709	Hexadecanoic acid, ethyl ester	1.604
25	26.31	<i>Trans</i> -delta(sup 9)-octadecenoic acid	0.153
26	26.776	Falcarinol	2.014
27	27.041	Oleic acid	1.666
28	27.132	9-octadecenoic acid (Z)-, eicosyl ester	0.930
29	27.403	Octadecanoic acid, methyl ester	1.992
30	28.283	3-methyl-2,6-dioxo-4-hexenoic acid	0.222
31	28.393	2-octadecoxyethanol	0.350
32	29.034	Delta 9- <i>cis</i> -oleic acid	0.664
33	30.276	Eicosane	0.430
34	30.502	Estra-1,3,5(10)-trien-17.β.-ol	2.932
35	32.656	1-(4-bromobutyl)-2-piperidinone	1.302
36	33.672	Venlafaxine	1.387
37	34.014	Butyl 9,12-octadecadienoate	5.228
38	37.171	7.7',8,8',11,11,12,12,15,15-decahydro psi-carotene	3.999
39	38.044	(2-fluorophenyl)methyl-1H-purin-6-amine	3.686
40	39.409	1.2.3.4-tetrol-L-ribo-octadecane	3.080
41	41.143	(6Z)-6-octadecenoic acid	1.299
42	41.46	9,10 dibromopentacosane	1.682
43	45.27	Glycerol 1-palmitate	4.536

low-fat Feta cheeses after 30 d of storage at 4°C. Table 5 shows that we observed no significant differences between all cheese samples in all parameters after 28 d

of storage, except for sample T3 which had the lowest ($P < 0.05$) color and appearance. The total score of the sensory evaluation is presented in Figure 2. Sample

Table 4. Antimicrobial activity $\pm$ SD of carrot powder methanolic extract

Bacterial strain ¹	Inhibition zone ² (mm)	
	Carrot powder methanolic extract	Cont
<i>Bacillus cereus</i> (positive) AUMC no. B-52	7.0 $\pm$ 2.0	20.0 $\pm$ 3.0
<i>Escherichia coli</i> (negative) AUMC no. B-53	8.0 $\pm$ 1.0	23.0 $\pm$ 3.0
<i>Micrococcus luteus</i> (positive) AUMC no. B-112	7.0 $\pm$ 3.0	22.0 $\pm$ 2.0
<i>Pseudomonas aeruginosa</i> (negative) AUMC no. B-73	10.0 $\pm$ 1.0	18.0 $\pm$ 3.0
<i>Serratia marcescens</i> (negative) AUMC no. B-55	8.0 $\pm$ 4.0	22.0 $\pm$ 2.0
<i>Staphylococcus aureus</i> (positive) AUMC no. B-54	7.0 $\pm$ 2.0	20.0 $\pm$ 3.0

¹AUMC no. = Assiut University Mycological Center.

²Cont = chloramphenicol as the antimicrobial standard.

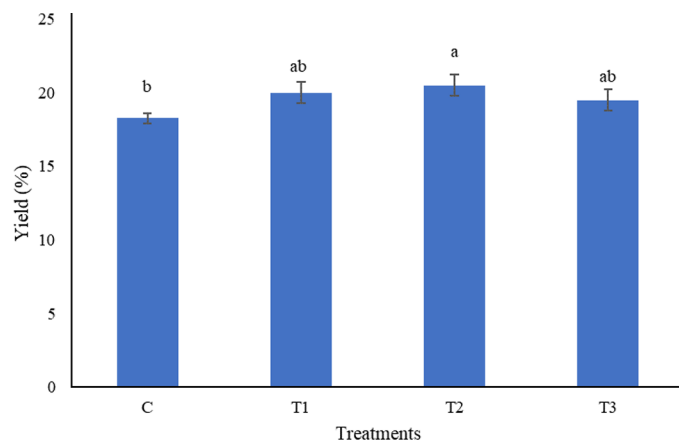


Figure 1. Cheese yield (%) of probiotic soft cheese supplemented with carrot powder. C = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. Means within treatments not sharing a common letter (a, b) are different ($P < 0.05$). Values shown are mean $\pm$ SD.

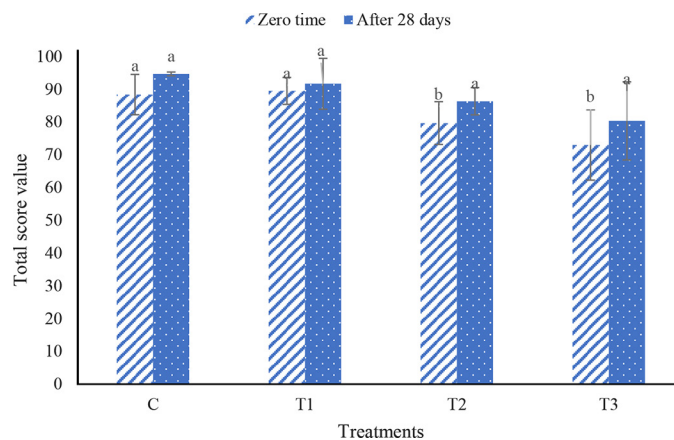


Figure 2. Total scores of organoleptic properties of probiotic soft cheese supplemented with carrot powder. C = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. Means within treatments not sharing a common letter (a, b) are different ($P < 0.05$). Values shown are mean $\pm$ SD.

T1 (probiotic cheese with 0.2% carrot powder) had the highest overall score ($P < 0.05$) at the beginning of the ripening, whereas we observed no significant differences in overall score between all cheese samples after 28 d of storage.

Protein Content and Nitrogenous Fractions.

The protein content of probiotic soft cheese is revealed in Table 6. The TP was calculated in fresh cheese samples by multiplying nitrogen content by 6.38. The TP was 24.84, 27.62, 27.67, and 31.26% in the control, T1, T2, and T3 cheese on a dry basis, respectively. These results reflected the enhancement of protein levels after carrot powder addition. It was found that the protein content in fresh soft cheeses made from cow milk ranged from 32.7 to 36.4% on a dry basis (Mohammed et al., 2016a). Table 6 illustrates that SN content in fresh con-

trol cheese samples was 0.6% (1.24% on a dry basis), which is in agreement with another study (Efthymiou and Mattick, 1964). The effect of rennet in casein hydrolysis was greater (into water-soluble peptides) than bacterial enzymes during the early stage of ripening (Pereira et al., 2010). Additionally, we observed a trend of decreasing SN values of fresh cheese samples at 0 d with an elevation in carrot powder concentration. This could be due to the effect of carrot powder on the rennet in the curd (keeping it trapped), referring to the curd ability to trap chymosin (McSweeney, 2004). In our study, at 28 d of ripening, SN content increased in all samples, while others found that SN contents in soft ultrafiltrate and control cheese samples at 21 d of ripening recorded higher values (1.41 and 1.08%, respectively; Furtado and Partridge, 1988). These dif-

Table 5. Sensory evaluation of probiotic soft cheese supplemented with carrot powder; values shown are mean $\pm$ SD

Storage period (d)	Treatment ¹	Flavor (50 points)	Body and texture (40 points)	Color and appearance (10 points)
0	Control	44.50 ^a $\pm$ 4.20	35.50 ^a $\pm$ 3.07	8.37 ^{ab} $\pm$ 1.18
	T1	44.87 ^a $\pm$ 3.83	36.00 ^a $\pm$ 2.26	8.62 ^a $\pm$ 0.91
	T2	40.87 ^{ab} $\pm$ 4.29	31.50 ^b $\pm$ 3.16	7.31 ^{bc} $\pm$ 0.79
	T3	36.37 ^b $\pm$ 5.85	29.87 ^b $\pm$ 5.43	6.75 ^c $\pm$ 1.48
28	Control	45.00 ^a $\pm$ 0.00	40.00 ^a $\pm$ 0.00	9.66 ^a $\pm$ 0.57
	T1	44.00 ^a $\pm$ 4.50	38.00 ^a $\pm$ 2.88	8.66 ^{ab} $\pm$ 0.57
	T2	40.00 ^a $\pm$ 1.15	37.00 ^a $\pm$ 2.51	8.00 ^{ab} $\pm$ 1.00
	T3	40.00 ^a $\pm$ 5.00	33.00 ^a $\pm$ 5.77	7.00 ^b $\pm$ 2.00

^{a-c}Means in the same column within the same period not sharing a common superscript are different ($P < 0.05$).

¹Treatment: control = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder.

Table 6. Protein content and nitrogenous fractions of probiotic soft cheese supplemented with carrot powder; values shown are mean $\pm$ SD

Storage period (d)	Treatment ¹	TP ²	SN	NPN
0	Control	24.84 ^c $\pm$ 0.00	0.60 ^a $\pm$ 0.00	0.60 ^a $\pm$ 0.00
	T1	27.62 ^b $\pm$ 0.51	0.41 ^b $\pm$ 0.06	0.38 ^b $\pm$ 0.00
	T2	27.67 ^b $\pm$ 0.96	0.35 ^b $\pm$ 0.073	0.27 ^d $\pm$ 0.00
	T3	31.26 ^a $\pm$ 0.99	0.40 ^b $\pm$ 0.02	0.36 ^c $\pm$ 0.00
28	Control		0.76 ^a $\pm$ 0.13	0.75 ^a $\pm$ 0.26
	T1		0.84 ^a $\pm$ 0.13	0.79 ^a $\pm$ 0.35
	T2		0.82 ^a $\pm$ 0.23	0.77 ^a $\pm$ 0.20
	T3		0.71 ^a $\pm$ 0.03	0.66 ^a $\pm$ 0.00

^{a-d}Means in the same column within the same period not sharing a common superscript are different ($P < 0.05$).

¹Treatment: control = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. TP = total protein; SN = soluble nitrogen.

²TP on a dry basis; TP = nitrogen content $\times$ 6.38.

ferences may be due to the difference in cheese salting, which delays the proteolysis activity (Park, 2001).

In contrast, NPN contents in control cheese samples at 0 d were higher than other cheese samples (Table 6), whereas Furtado and Partridge (1988) found that NPN values after 1 d of ripening were 0.30 and 0.21% in soft ultrafiltrate and control cheese samples, respectively. The NPN values revealed the capability of microbial origin enzymes to degrade large peptides (which are released by the rennet) and produced small peptides, free amino acids, and ammonia nitrogen (Calvo et al., 2007). Table 6 shows that the addition of carrot powder contributed to secondary proteolysis after 28 d of storage; however, this addition decreased the NPN nonsignificantly. These results may be attributed to the inhibition effect of carrot powder on proteolytic bacteria.

Figure 3 shows the ripening extension index in cheese made with carrot powder. The ripening extension index ranged from 25.41 to 36.58% at 0 d, with the control sample being the highest ($P < 0.05$) in the ripening extension index. Similar results were reported by Efthymiou and Mattick (1964). After 28 d of ripening, the extension index increased at a slow rate, which might be attributed to the low temperature during storage, and this, in turn, reduce the rate of protein hydrolysis (Daigle et al., 1999). Furthermore, Furtado and Partridge (1988) found that the ripening extension index values in soft ultrafiltrate and control cheese samples after 21 d of ripening were 48.5 and 41.1%, respectively.

However, Figure 4 shows the ripening depth index in all cheese samples at 0 d and after 28 d of storage. Lower values of the ripening depth index were found by Furtado and Partridge (1988). These differences might be attributed to the use of 4 bacterial starters with rennet in the current study, which activates casein hydrolysis (Sousa et al., 2001). Furthermore, the addi-

tion of carrot powder caused a decrease in the ripening depth index. Although we observed the ability of carrot addition to increase the numbers of LAB (which release enzymes and produce NPN), this addition had a small influence on proteolysis due to the low ability of *Lactiplantibacillus plantarum* ssp. *plantarum* in protein hydrolysis (Zhang et al., 2021). In addition, LAB consume amino acids as a nitrogen source during the growth (Pereira et al., 2010).

Moisture, Fat, Salt, and Acidity Contents.

Table 7 exemplifies that the moisture content of probiotic cheese supplemented with carrot powder increased significantly ($P < 0.05$) by elevating the concentration of carrot addition. This increase could be due to the enhancement of water holding capacity (Guyih et al., 2020). We observed a trend in decreasing water content during storage so the control sample had the lowest moisture content (51.66%) after 28 d of storage. Similar results were noticed by others, as they found that the moisture content decreased in cheese samples during storage (Abd El-Rafee and Abd El-Gawad, 2002). In contrast, Table 7 reveals that cheeses treated with carrot powder had higher moisture contents and lower fat (especially in sample T3). Similar findings were reported by other researchers when they treated soft white cheese with cranberry fruit extract (Khalifa and Wahdan, 2015). Meanwhile, it was found that fat content ranged between 25.5 and 25.75% in probiotic cheeses with different starters (Yerlikaya and Ozer, 2014). After 28 d of storage, fat content increased in cheese treated with carrot powder (Table 7) due to the increment of dry matter. Elevating the content of carrot powder to 0.6% during the making of probiotic soft cheese had a significant effect ($P < 0.05$) on salt contents, as sample T3 recorded the highest ash value (4.09%). These results might be related to the increment of moisture content with carrot addition, which increases the cheese's

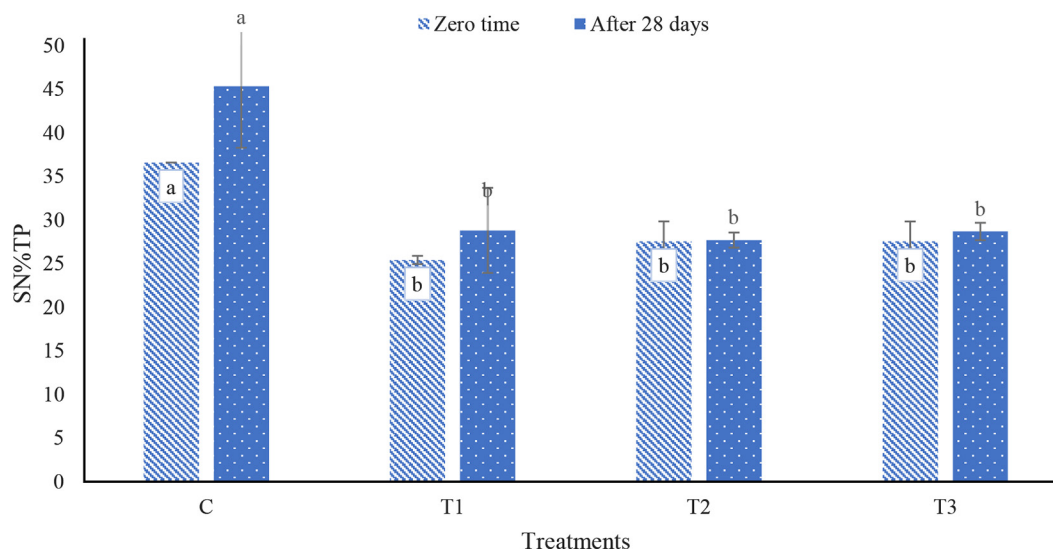


Figure 3. Ripening extension index of probiotic soft cheese supplemented with carrot powder. SN%TP = soluble nitrogen as a percentage of total protein; C = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. Means within treatments not sharing a common letter (a, b) are different ($P < 0.05$). Values shown are mean $\pm$ SD.

ability to absorb more salt (Ayyash et al., 2018). Moreover, Yerlikaya and Ozer (2014) found that salt content ranged from 4.91 to 5.03% in white cheeses, which were manufactured with different probiotic bacteria. Even though throughout the ripening period, salt content fluctuated due to the diffusion of salt between whey and cheese samples (Topçu and Saldamli, 2006; Pereira et al., 2010), we observed no significant differences ($P > 0.05$) between all fresh cheese samples in titratable acidity. By increasing the storage period, the control cheese samples had a higher significant ($P < 0.05$) ti-

tratable acidity than the cheese samples treated with carrot powder. These results agreed with the findings of several studies (Yerlikaya and Ozer, 2014; Khalifa and Wahdan, 2015) as they found a similar trend in the titratable acidity.

Microbiological Characteristics. The TBC of probiotic soft cheese made with carrot is shown in Table 8. We observed an increase in TBC during storage. Similarly, Khalifa and Wahdan (2015) reported a rise in TBC in soft white cheese during the storage period. However, our study showed a significant reduc-

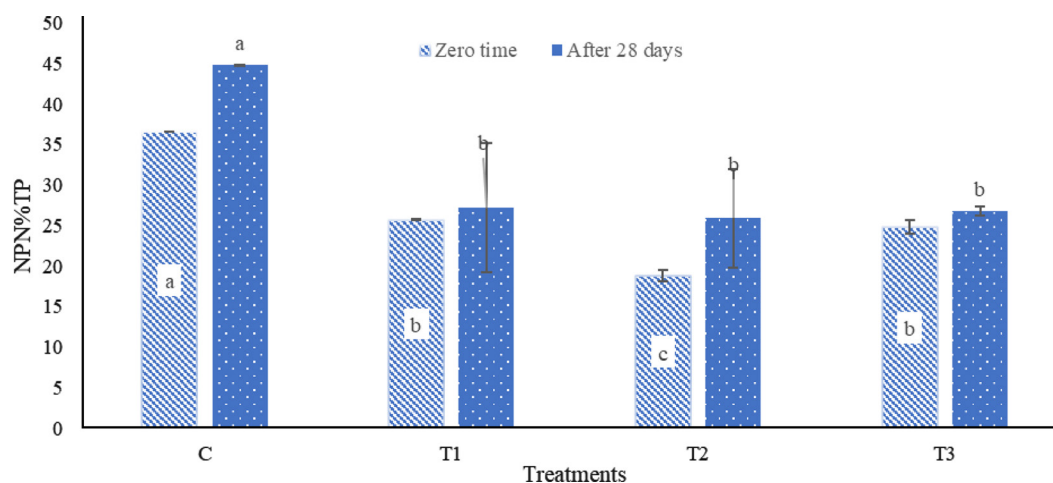


Figure 4. Ripening depth index of probiotic soft cheese samples supplemented with carrot powder. NPN%TP = nonprotein nitrogen as a percentage of total protein; C = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. Means within treatments not sharing a common letter (a, b) are different ($P < 0.05$). Values shown are mean $\pm$ SD.

Table 7. Mean ($n = 3$) $\pm$ SD composition (%) of probiotic soft cheese supplemented with carrot powder during storage for 28 d

Item	Treatment ¹	Storage period (d)					Mean
		0	7	14	21	28	
Moisture	Control	57.88 $\pm$ 0.88	58.65 $\pm$ 0.83	66.26 $\pm$ 3.85	59.66 $\pm$ 1.08	51.66 $\pm$ 0.60	58.82 ^c
	T1	66.18 $\pm$ 0.77	62.03 $\pm$ 5.88	66.30 $\pm$ 1.61	58.94 $\pm$ 2.81	61.43 $\pm$ 3.33	62.97 ^b
	T2	66.72 $\pm$ 0.72	62.00 $\pm$ 3.42	68.30 $\pm$ 1.35	65.84 $\pm$ 1.02	59.51 $\pm$ 0.30	64.47 ^b
	T3	70.61 $\pm$ 0.61	67.26 $\pm$ 1.76	68.18 $\pm$ 1.23	66.70 $\pm$ 1.07	67.60 $\pm$ 1.62	68.07 ^a
	Mean	65.34 ^B	62.48 ^C	67.26 ^A	62.78 ^C	60.05 ^D	
Fat	Control	24.00 $\pm$ 1.00	24.66 $\pm$ 0.57	25.33 $\pm$ 1.15	20.66 $\pm$ 0.57	20.66 $\pm$ 0.57	23.06 ^b
	T1	23.33 $\pm$ 1.52	21.00 $\pm$ 1.00	23.66 $\pm$ 2.51	24.66 $\pm$ 0.57	31.33 $\pm$ 1.15	24.80 ^a
	T2	22.00 $\pm$ 2.64	19.66 $\pm$ 0.57	22.66 $\pm$ 3.21	22.33 $\pm$ 0.57	28.33 $\pm$ 0.57	23.00 ^b
	T3	21.00 $\pm$ 1.15	17.33 $\pm$ 0.57	23.66 $\pm$ 0.57	20.33 $\pm$ 1.15	23.33 $\pm$ 0.05	21.20 ^c
	Mean	22.66 ^C	20.66 ^D	23.83 ^B	22.00 ^C	25.92 ^A	
Salt	Control	3.97 $\pm$ 0.04	4.16 $\pm$ 0.00	4.19 $\pm$ 0.16	3.60 $\pm$ 0.04	3.77 $\pm$ 0.04	3.94 ^{bc}
	T1	3.65 $\pm$ 0.00	4.31 $\pm$ 0.01	4.18 $\pm$ 0.04	3.75 $\pm$ 0.04	3.65 $\pm$ 0.00	3.91 ^c
	T2	3.99 $\pm$ 0.08	4.16 $\pm$ 0.00	3.77 $\pm$ 0.04	3.99 $\pm$ 0.04	3.85 $\pm$ 0.04	3.95 ^b
	T3	4.09 $\pm$ 0.00	4.53 $\pm$ 0.00	4.01 $\pm$ 0.07	3.92 $\pm$ 0.04	3.71 $\pm$ 0.01	4.05 ^a
	Mean	3.93 ^C	4.29 ^A	4.04 ^B	3.82 ^D	3.75 ^E	
Acidity	Control	0.45 $\pm$ 0.00	0.45 $\pm$ 0.00	0.90 $\pm$ 0.0000	0.90 $\pm$ 0.0	1.15 $\pm$ 0.05	0.77 ^a
	T1	0.52 $\pm$ 0.12	0.67 $\pm$ 0.00	0.67 $\pm$ 0.00289	0.75 $\pm$ 0.12	0.90 $\pm$ 0.00	0.70 ^b
	T2	0.45 $\pm$ 0.00	0.67 $\pm$ 0.00	0.67 $\pm$ 0.0050	0.67 $\pm$ 0.00	0.67 $\pm$ 0.00	0.62 ^c
	T3	0.45 $\pm$ 0.00	0.67 $\pm$ 0.00	0.67 $\pm$ 0.0050	0.67 $\pm$ 0.00	0.45 $\pm$ 0.00	0.58 ^d
	Mean	0.46 ^D	0.61 ^C	0.73 ^B	0.75 ^B	0.79 ^A	

^{a-d}Means in the same column within the same period not sharing a common superscript are different ($P < 0.05$).

^{A-D}Means in the same row within the same period not sharing a common superscript are different ($P < 0.05$).

¹Treatment: control = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder.

tion ($P < 0.05$) of TBC at 0 d of storage for samples T2 and T3 containing carrot concentrations of 0.4 and 0.6%, respectively, compared with the control sample. The decline of TBC in cheese made with carrot powder revealed the preservative effect of carrot used in probiotic soft cheese. The current study also evaluated the growth of LAB and *Bifidobacterium* species during the storage period (Table 8) and observed a significant ($P < 0.05$) increase in both probiotic cultures during the storage of samples T2 and T3. Because LAB are known to release bacteriocins, the reduction in TBC counts can be a result of the increase in LAB (Arqués et al., 2015; Garnier et al., 2019). Lactic acid bacteria recorded their highest number in sample T3 after 21 d of storage, followed by a decrease from 8.77 to 8.47 log cfu/g (Table 8). This decline may be due to the consumption of nutrients and lactic acid production by LAB, which affected their growth and numbers (Mushtaq et al., 2015). A previous study referred to the recommended level of probiotic bacteria in food during consumption as 6 log cfu/g (Karimi et al., 2012), whereas in our study, LAB and *Bifidobacterium longum* counts were 8.47 and 9.40 log cfu/g in T3 after 28 d of ripening. These results indicate that probiotic bacteria can survive in the final cheese with carrot powder addition during ripening, as carrot powder induced and

maintained the growth of probiotic bacteria. However, proteolytic bacteria appeared in all cheese samples after 14 d of storage, followed by a decrease in counts by the end of storage. Similar growth trends of proteolytic and lipolytic bacteria were reported (Khalifa and Wahdan, 2015). Furthermore, elevating the carrot powder concentration in probiotic cheese samples resulted in a decrease in proteolytic counts, while no lipolytic bacteria were observed. However, psychotropic counts were observed after 28 d of storage for all samples. Test T3 recorded the lowest psychotropic count. The results of yeast and mold analysis revealed the growth of molds in the control samples only after 21 d of storage, whereas yeast counts were not detected in any of the cheese samples during storage for 28 d.

CONCLUSIONS

Results of the present study indicated that carrot powder is a good source of nutrients, such as minerals, β -carotene, lycopene, phenolic compounds, and flavonoids. We also found that carrot powder enhanced the antimicrobial and antioxidative value of the probiotic cheese. Using carrot powder as a functional ingredient in probiotic soft cheese induces the growth of probiotic bacteria, and therefore can be used as a

Table 8. Mean ($n = 3$) $\pm$ SD microbiological characteristics (log cfu/g) of probiotic soft cheese supplemented with carrot powder during storage for 28 d

Item	Treatment ¹	Storage period (d)					Mean
		0	7	14	21	28	
Total bacterial count (TBC)	Control	7.48 $\pm$ 0.04	8.30 $\pm$ 0.02	9.38 $\pm$ 0.01	9.44 $\pm$ 0.01	9.29 $\pm$ 0.06	8.78 ^a
	T1	7.47 $\pm$ 0.040	8.25 $\pm$ 0.05	9.29 $\pm$ 0.02	9.35 $\pm$ 0.00	9.29 $\pm$ 0.04	8.73 ^b
	T2	7.37 $\pm$ 0.015	8.23 $\pm$ 0.01	9.17 $\pm$ 0.01	9.19 $\pm$ 0.00	8.99 $\pm$ 0.02	8.59 ^c
	T3	7.35 $\pm$ 0.04	7.71 $\pm$ 0.02	7.99 $\pm$ 0.08	8.05 $\pm$ 0.05	7.99 $\pm$ 0.04	7.81 ^d
	Mean	7.42 ^E	8.12 ^D	8.96 ^B	9.01 ^A	8.89 ^C	
Lactic acid bacteria (LAB)	Control	7.11 $\pm$ 0.06	7.64 $\pm$ 0.02	7.59 $\pm$ 0.04	6.57 $\pm$ 0.03	6.47 $\pm$ 0.01	7.07 ^d
	T1	7.13 $\pm$ 0.05	7.72 $\pm$ 0.02	7.99 $\pm$ 0.04	7.47 $\pm$ 0.01	7.47 $\pm$ 0.03	7.55 ^c
	T2	7.14 $\pm$ 0.03	7.80 $\pm$ 0.03	8.17 $\pm$ 0.02	8.46 $\pm$ 0.11	8.29 $\pm$ 0.02	7.97 ^b
	T3	7.17 $\pm$ 0.03	7.90 $\pm$ 0.05	8.36 $\pm$ 0.01	8.77 $\pm$ 0.03	8.47 $\pm$ 0.07	8.13 ^a
	Mean	7.13 ^E	7.76 ^C	8.03 ^A	7.82 ^B	7.67 ^D	
<i>Bifidobacterium longum</i>	Control	7.65 $\pm$ 0.04	7.49 $\pm$ 0.11	7.20 $\pm$ 0.01	6.84 $\pm$ 0.01	6.71 $\pm$ 0.02	7.17 ^c
	T1	7.70 $\pm$ 0.01	8.29 $\pm$ 0.02	9.14 $\pm$ 0.12	9.09 $\pm$ 0.33	8.90 $\pm$ 0.00	8.62 ^b
	T2	8.18 $\pm$ 0.65	8.40 $\pm$ 0.01	9.38 $\pm$ 0.16	9.19 $\pm$ 0.05	9.15 $\pm$ 0.02	8.86 ^a
	T3	8.79 $\pm$ 0.91	8.45 $\pm$ 0.04	9.39 $\pm$ 0.03	9.31 $\pm$ 0.34	9.40 $\pm$ 0.27	9.07 ^a
	Mean	8.08 ^B	8.16 ^B	8.78 ^A	8.60 ^A	8.54 ^A	
Proteolytic	Control	ND	ND	7.55 $\pm$ 0.01	8.31 $\pm$ 0.01	6.94 $\pm$ 0.14	7.60 ^a
	T1	ND	ND	7.49 $\pm$ 0.02	7.92 $\pm$ 0.03	6.71 $\pm$ 0.80	7.37 ^a
	T2	ND	ND	7.27 $\pm$ 0.03	6.31 $\pm$ 0.01	6.44 $\pm$ 1.18	6.67 ^b
	T3	ND	ND	6.74 $\pm$ 0.04	6.80 $\pm$ 0.28	6.73 $\pm$ 0.77	6.75 ^b
	Mean			7.26 ^A	7.33 ^A	6.70 ^B	
Psychotropics	Control	ND	ND	ND	ND	8.26 ^a	
	T1	ND	ND	ND	ND	8.27 ^a	
	T2	ND	ND	ND	ND	8.20 ^b	
	T3	ND	ND	ND	ND	7.73 ^c	
	Mean						
Lipolytic	Control	ND	ND	ND	6.54	6.3	
	T1	ND	ND	ND	ND	ND	
	T2	ND	ND	ND	ND	ND	
	T3	ND	ND	ND	ND	ND	
	Mean						
Mold	Control	ND	ND	ND	6	6.89	
	T1	ND	ND	ND	ND	ND	
	T2	ND	ND	ND	ND	ND	
	T3	ND	ND	ND	ND	ND	
	Mean						
Yeast	Control	ND	ND	ND	ND	ND	
	T1	ND	ND	ND	ND	ND	
	T2	ND	ND	ND	ND	ND	
	T3	ND	ND	ND	ND	ND	
	Mean						

^{a-d}Means in the same column within the same period not sharing a common superscript are different ($P < 0.05$).

^{A-D}Means in the same row within the same period not sharing a common superscript are different ($P < 0.05$).

¹Treatment: control = probiotic soft cheese without any carrot; T1 = probiotic soft cheese with 0.2% carrot powder; T2 = probiotic soft cheese with 0.4% carrot powder; T3 = probiotic soft cheese with 0.6% carrot powder. ND = not detected.

potential prebiotic to improve the viability of probiotic bacteria in various food applications similar to white cheese.

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