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Enhancing The Nutritional Value and Chemical Composition of Functional Yogurt Drink by Adding Bee Honey and *Spirulina* Powder

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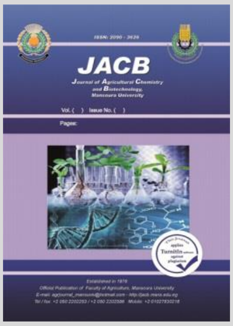

ABSTRACT

Recent societal interest in healthful food has led to the development of functional dairy products that primarily provide health benefits in addition to their essential nutrients. The main objective of the research is to extract, estimate, and fractionation the bio-active and antioxidant compounds and pigments in *Spirulina* algae and apply their use in a functional yogurt drink that enhances nutritional value by adding pure bee honey to it to give it shape, taste and high nutritional value and replacement of cane sugar with natural honey. *Spirulina* is added to yogurt drinks at concentrations of 0.5, 1, and 1.5 percent with traditional starters and 0.5, 1, and 1.5 percent with traditional starters plus *Bifidobacterium longum*. T.S.S, α-Tocopherol, and ash content were determined. With an increase in *Spirulina* concentration, a decrease in pH was seen during the setting period. Diacetyl, Acetoin, antioxidants, and phytochemicals all increased along with the level of *Spirulina*. Yogurt drinks made with 1% *Spirulina* scored similarly to controls for sensory factors like color, and appearance, flavor, and texture. All Yogurt drink samples were mold and yeast free. Liveliness of the Yogurt drink culture when stored at the recommended temperature of 5°C: Control has less overall lactic acid bacteria, but all samples can increase their *Spirulina* content by 0.5, 1, and 1.5 times as well as their overall bifidobacterial count the progress of storage for all lactic acid bacteria and bifidobacterial up to 15 days the viable count.

***Keywords**:* Spirulina powder, Phycocyanin pigment, and yogurt drink.

INTRODUCTION

Spirulina has a high calcium and iron content (1043.62 or 338.76 mg/100 g), making it excellent for children's growth. Additionally, it contains plant pigments (phycocyanin: 14.1% and chlorophyll: 1.4%), as well as selenium (0.0488 mg/100 g). These items include powerful antioxidants, physical chemistry, sensory properties, fresh *Spirulina*, and yogurt. The *Spirulina* cell wall is made up of protein, carbohydrates, and fat, it should be noted. Therefore, compared to other food sources, particularly plant food sources, *Spirulina* may have a higher bioavailability of nutrients. *Spirulina* has more protein than any other natural food (62 %). The Total flavonoid can be found in *Spirulina* It is the richest natural source of vitamin E, Phycocyanin, and carotene. (Ali *et al.*, 2022, Hassan *et al.*, 2021 and Darwish *et al.*, 2020).

Spirulina can therefore ensure that food is whole and that the body is in a balanced alkaline state. *Spirulina* has a variety of positive health effects because of its chemical makeup. Many diseases, including cancer, kidney failure, and high blood pressure, are slowed down by it (Danesi *et al.*, 2010).

Contrarily, the characteristics of *Spirulina* depend on a variety of factors, including B. its characteristics, raw materials, pretreatment, processing, and storage. *Spirulina* contains several bioactive substances, some of which are heat sensitive. Processing technology must therefore be strictly adhered to (Desmorieux and Hernandez, 2004) demonstrated the reduction in protein and total sugar content following convection drying and oven drying with infrared. The physical-chemical, nutritional, rheological, sensory, and technical functional properties of *Spirulina* may change as a result of these treatments. This *Spirulina*'s qualities differ depending on whether it is processed or fresh.

Humans frequently consume *Spirulina* in a variety of forms, including tablets, capsules, and food additives. In the same way that candies, and gel desserts (Guarda *et al.*, 2004), baked goods (Gouveia *et al.*, 2008), sweets (Varga *et al.*, 2002), and dairy products (Wherry *et al.*, 2019). *Spirulina* is currently added to dairy products for human nutrition because of its wide-ranging nutritional value. Protein, minerals, and water-soluble vitamins are beneficial. Felfoul *et al.*, (2017) found that yogurt was a staple in the diet of the majority of the population. These products' fortification becomes effective. Reduce illnesses brought on by dietary deficiencies. However, the current industry and researchers never use fresh shapes of *Spirulina*; only dried powder.

People can use probiotics as healthy sources in their diets as food supplements. They can be added to many different foods to boost and provide health benefits (Rybka and Kailasapathy, 1995). Many health benefits are associated with the majority of commercially available probiotics, such as *Lactobacillus* and *Bifidobacterium* (Mortazavian *et al.*, 2005). Additionally, they are effective in supplying the host with health benefits (FAO/WHO, 2001), including the development of lactose tolerance (Kim and Gilliland, 1983),

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antimicrobial (Yildirim and Johnson, 1998), anticarcinogenic (El-Gawad *et al.,* 2004), hypercholesterolemic (Kikuchi-Hayakawa *et al.,* 2000), and antimutagenic (Hsieh and Chou, 2006) by growing its intestinal microbial scales (Molna'r *et al.,* 2009). Additionally, the sensory qualities of final yields will be impacted by *Spirulina* injections in fermented milk. Functional foods are those that have a high concentration of bioactive compounds like alkaloids, carotenoids, flavonoids, phenolic acids, and compounds, stilbenes, and lignans, tannins, terpenes, that are used for specific therapeutic purposes or human health. as well as terpenoids (Alongi and Anese, 2021 and Banwo *et al.,* 2021).

According to a recent review by Macori and Cotter (2018), the major and minor milk components are converted through controlled microbial culturing and enzymatic conversion to produce the fermented dairy products we consume today. Increased microbiological safety, longer shelf life, flavor addition, and improved palatability and organoleptic qualities are all benefits of fermentation. Microorganisms carry out a series of intricate chemical reactions during the Milk undergoes a fermentation process that creates new molecules with improved nutritional value and digestibility. Additionally, fermentation produces metabolites that can significantly contribute to a daily healthy diet (Marco *et al.,* 2017).

Due to their high lipid, protein, carbohydrate, and mineral content, fermented foods have been consumed for centuries. fermented foods are highly nutritious. It may reduce the risk of colorectal cancer and alleviate diarrhea, among other health benefits. (Saad *et al.,* 2013 and Pala *et al.,* 2011) These health advantages are believed to be mediated by preserving a proper balance of the gut microbiota and controlling the intestinal immune system's microenvironment. (Kok and Hutkins, 2018) and (Fernandez *et al.,* 2017)

Yogurt is thought to contain significant amounts of bioactive peptides, which are produced during fermentation. As a result, numerous attempts to produce yogurt that has been fortified with natural antioxidants and presents a novel product has generated a lot of interest (Caleja *et al.,* 2016, Gahruie *et al.,* 2015). Yogurt is a popular beverage for cooling because of its low viscosity. One of the fermented milk products with the fastest global growth is yogurt drink, also known as drinkable yogurt (Douaud, 2007, Ho *et al.,* 2000). This popular fermented beverage is typically made from milk, yogurt, or drinking yogurt. It helps treat lactose intolerance, prevents diarrhea, lowers blood cholesterol, and lowers the risk of digestive system tumors or cancer, among other things (Legowo, 2009).

Honey is a supersaturated solution of fructose and glucose (Gheldof and Engeseth, 2002) and contains trace components such as phenolic acids and flavonoids, glucose oxidase and catalase, ascorbic acid, carotenoids, organic acids, amino acids, proteins and Alpha-tocopherol (Ferreres *et al.,* 1993). Honey is bacteriostatic and bactericidal against Gram-positive bacteria (Bogdanov, 1997), and antimicrobial activity has been attributed to peroxide and non-peroxide substances.

As consumers become more conscious of healthy foods, the basic concept of nutrition is undergoing major changes. Foods can no longer be judged solely on the basis of their macronutrient and micronutrient content. Consumers believe that certain foods can demonstrate health benefits, which has led to the term functional food, consumers are increasingly concerned about the health benefits of functional products, thereby driving the growth of the global dairy market. Functional foods are a new direction in science, somewhere between food and medicine, and thus have great potential to improve health and prevent disease when consumed as part of a balanced diet. The recent societal interest in healthy foods has led to the development of functional dairy products that provide health benefits in addition to providing essential nutrients (Sarkar, 2019).

There is well-documented evidence that products containing probiotics have health benefits, and consumer demand for natural products has also fueled consumer preferences for probiotic foods (Sarkar, 2018). Probiotics can improve human health by altering the gut microbiota to affect physiological and pathological processes in the host (Sarkar *et al.,* 2016). Due to their therapeutic and nutritional properties, probiotics have now become one of the most powerful functional foods and have been shown to be effective in modulating the gastrointestinal flora to prevent the spread of various diseases (Sarkar *et al.,* 2017). Sanders and Marco (2010) and Codex Standard, (2001) reported that probiotics grow better in food matrices containing bioactive ingredients such as prebiotics, vitamins, minerals, fibers, enzymes, food preservatives, and flavorings.

The main objective of the research is to extract, estimate, and fraction the bio-active and antioxidant compounds and pigments in *Spirulina* algae and apply their use in a functional yogurt drink that enhances nutritional value by adding pure bee honey to it to give it shape, taste and high nutritional value and replacement of cane sugar with natural honey and to assess the sensory and textural properties of the developed functional yogurt drink.

MATERIALS AND METHODS

Milk samples: At Minia University's Department of Animal Production in the Faculty of Agriculture, all fresh cow's milk samples were taken from the herd.

Starters: Yogurt starter consisted of *Loctobacillus delbrueckii* subsp. *bulgaricus* (EMCC 11102), *Streptococcus salivarius* subsp. *Thermophilus* (EMCC 11044) and *Bifidobacterium longum* (ATCC 15707) was obtained from Cairo Microbiological Resource's Center (MIRCEN) Faculty of Agriculture, Ain Shams University.

A sample of clover honey was obtained from the apiary of the Faculty of Agriculture, Minia University, and it is a completely natural honey that was produced from the nectar of clover flowers without feeding the bees at all.

Preparation of yogurt drink:

Egyptian cow's milk was heated at 90 Cº for 5 min. The milk was cooled to 45EC and then honey (8.5%) was dissolved in the milk and it was divided into 2 portions. The first potion was inoculated with a starter culture (DVS Express 1) and the second potion was inoculated with a probiotic starter culture. Six treatments were carried out as follows C; yogurt culture with add 10% of boiled water as a control, T1; yogurt culture with adding 0.5% *Spirulina* powder and 10% of boiled water, T2; yogurt culture with adding 1% *Spirulina* powder and 10% of boiled water, T3; yogurt culture with adding 1.5% *Spirulina* powder and 10% of boiled water; T4; yogurt culture and bifidobacterial culture with adding 0.5% *Spirulina* powder and 10% of boiled water, T5; yogurt culture and bifidobacterial culture with adding 1% *Spirulina* powder and 10% of boiled water, T6; yogurt culture and bifidobacterial culture with adding 1.5% *Spirulina* powder and 10% of boiled water. Treatments were incubated at 42EC until pH drops to 4.7 and then kept at 5 EC overnight. Coagulated yogurt was stirred by a mechanical mixer, filled in plastic bottles, and stored at 5 EC for 15 days.

Chemical composition of sample:

Physiochemical parameters: of total solids, Moisture, ash, pH, and protein contents of drinking yogurt with *Spirulina* powder were determined by AOAC, (2007).

Tocopherol analysis: The total tocopherol content in the sample was determined according to the method of Wong *et al.,* (1988).

Total phenolic content: was determined according to o Shiri *et al.,* (2011).

Total flavonoid content: Total flavonoid content was determined according to (Zilic *et al.,* 2012).

Color characteristics were measured by a color difference meter (model color Tec-PCM, USA) Wallace and Giusti (2008). Where: L: Value represents darkness from black (0) to white (100) a: Value represents color ranging from red (+) to green (-) b: Value represents yellow (+) to blue.

microbiological analysis:

For the total bacterial count, standard plate count agar was utilized (ISO, 2013). Total lactic acid bacteria were counted using MRS agar, and total bifidobacteria were counted using MRS+ L cysteine agar (Dave and Shah, 1996). The counting of yeasts and molds was done following (ISO, 2008).

Sensory analysis: A sensorial rating scale of 10 points for texture, 10 points for color, 20 points for flavor, and 40 points for acceptability was used by a panel of 10 trained panelists to assess the sensory qualities of the drinking yogurt samples.

Statistic evaluation of the means and standard deviations (SD) of three measurements represented the experimental results. ANOVA procedures were used to perform the analysis of variance. For statistical calculations, GraphPad Prism® was used. US company Graph Pad Software, San Diego, CA (Motulsky, 1999).

RESULTS AND DISCUSSION

Tables provide a summary of the dried powdered *Spirulina*'s chemical and nutritional makeup (1, 2, and 3). The results of the chemical composition of *Spirulina* are in agreement with those obtained by Branger *et al.,* (2003); Habib *et al.,* (2008); Vijayarani *et al.,* (2012), Dolly, (2014), and Sharoba, (2014).

Table 1. Chemical composition (%) of *Spirulina* algae powder.

Ingredients (%)	Spirulina algae
Moisture	4.75±0.05
Protein	62.00±0.08
Fat	8.50±0.07
Fiber	6.49±0.05
Ash	11.40±0.03
Carbohydrates	13.80±0.07

Data are expressed as mean ± SD values given to represent the means of three determinations.

Table 2. Phytochemical composition in *Spirulina* algae(mg/100gm).

Phytochemicals	Spirulina algae
Tocopherols (ppm)	250.00±0.33
Total polyphenols (ppm)	300.00±0.45
Total flavonoid (ppm)	817.00±0.55

Data are expressed as mean ± SD values given to represent the means of three determinations.

Table 3. Pigments content in *Spirulina* algae (mg/100gm).

Pigments	Spirulina algae
Carotene	15.00±0.04
Xanthophylls	0.90±0.01
Zeaxanthin	0.30±0.03
Chlorophyll	25.00±0.05
Phycocyanin	327.00±0.35

Data are expressed as mean ± SD values given to represent the means of three determinations.

Moisture content of honey is a limiting factor in determination of its quality (Table 4), stability and spoilage resistance against yeast fermentation. The higher the moisture content is the higher probability of honey fermentation during storage. Lower moisture limits (<20%), elongates honey shelf life which would be met by a large majority of the commercial honeys, have been proposed by some countries for the revision of the (Codex Alimentations, 2001). These results were accepted by the international regulations for honey quality. Clover honey recorded moisture contents (21.66 %), 78.34 T.S.S these results are consistent with Iryna *et al.,* 2001 and it contains 3.953 ASH Generally speaking, honey has a low ash content, which is influenced by the nectar's chemical makeup varies depending on the various botanical sources used in the production of honey (Andrade *et al.,* 1999). The range is 0.02 to 1.0%, and the maximum permitted by law for honey derived from floral sources is 0.6 % (Codex Alimentarius, 1993). However, this honey and terpenoids typically have ash contents between 0.1 and 0.3% (Codex Alimentarius, 2000), Honey contains about 0.5% proteins according to (Bogdanov *et al.,* 2008).

Table 4. Chemical composition of honey

	Moisture	T.S.S	Ash	Proteins
Honey	21.66±0.2417	78.34±0.3051	0.65±0.04726	0.5 ±0.04726

Data are expressed as mean ± SD values given represent means of three determinations

Chemical composition of supplemental yogurt drink:

The effect of enrichment of yogurt drink with *Spirulina* on the gross composition of yogurt drink (Table 5) indicated that with an increase in enrichment level from 0 to 1.5 percent, there was a significant increase in protein from 3.803 to 7.090 percent, total solids from 19.64 to 24.32 percent. It is well known that *Spirulina* is a great source of protein. The inherent composition of *Spirulina* may be to blame for the compositional change of yogurt beverages. However, the *Spirulina*-enriched yogurt drink's increased ash content was determined to be significant, and these findings coincided with Priyanka *et al.,* (2013).

The pH of supplemental yogurt drinks:

According to the findings, increasing the amount of *Spirulina* from 0.5% to 1.5 % resulted in a drop in pH from 4.785 to 3.60 (T3) and from 4.785 to 3.195 (T6). According to Varga *et al.,* (2002), Perez *et al.,* (2007), and Priyanka *et al.,* (2013), *Spirulina* addition significantly changes the pH of yogurt drinks when compared to the control. *Spirulina* biomass has an alkaline nature and is capable of accelerating the growth of dairy starter culture while also producing acid during fermentation.

Table 5. Chemical composition of supplemental yogurt drink with *Spirulina* powder

		Moisture	T.S.S	Ash	Proteins
C	Fresh	80.36±0.2	19.64±0.4454	0.6600±0.02646	3.603±0.02517
	7 days	80.21±0.3	19.79±0.4650	0.7033±0.01528	3.790±0.04583
	15 days	80.14±0.13	19.86±0.2646	0.7173±0.006429	3.840±0.1311
T1	Fresh	79.79±0.3	20.21±0.2281	0.8257±0.01365	5.323±0.02517
	7 days	79.58±0.5	20.42±0.1419	0.8517±0.007638	5.423±0.02082
	15 days	79.36±0.5	20.64±0.1528	0.8773±0.01026	5.600±0.1562
T2	Fresh	79.58±0.4994	20.42±0.3523	0.9310±0.008544	6.723±0.02517
	7 days	79.52±0.5499	20.48±0.1041	0.9657±0.006028	6.747±0.04509
	15 days	79.22±0.2250	20.78±0.3045	0.9860±0.009644	6.817±0.01528
T3	Fresh	75.68±0.5773	24.32±0.1704	1.070±0.04583	6.883±0.09452
	7 days	75.64±0.1353	24.36±0.1861	1.507±0.08021	7.050±0.04583
	15 days	75.47±0.4339	24.53±0.1790	1.523±0.06658	7.090±0.03606
T4	Fresh	79.80±0.3372	20.20±0.09644	0.8453±0.01550	5.433±0.04163
	7 days	79.69±0.2902	20.310±0.1002	0.8733±0.01528	5.570±0.1136
	15 days	79.64±0.2946	20.36±0.1229	0.8700±0.01000	5.640±0.09165
T5	Fresh	79.14±0.5168	20.20±0.1572	0.9800±0.01000	6.663±0.09074
	7 days	79.06±0.03786	20.94±0.2616	0.9803±0.009504	6.717±0.01102
	15 days	78.53±0.4709	21.47±0.2801	1.001±0.01762	6.833±0.03055
T6	Fresh	75.70±0.2346	24.30±0.3055	1.110±0.03606	7.053±0.03215
	7 days	75.70±0.3000	24.30±0.1572	1.470±0.1353	7.077±0.01528
	15 days	75.67±0.1528	24.33±0.1253	1.810±0.1572	7.113±0.01528

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05 C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacteria culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacteria culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacteria culture with adding 1.5% Spirulina.

The pH of supplemental yogurt drinks:

According to the findings, increasing the amount of *Spirulina* from 0.5% to 1.5 % resulted in a drop in pH from 4.785 to 3.60 (T3) and from 4.785 to 3.195 (T6) (Table 6). According to Varga *et al.*, (2002), Perez *et al.*, (2007), and Priyanka *et al.*, (2013), *Spirulina* addition significantly changes the pH of yogurt drinks when compared to the control. *Spirulina* biomass has an alkaline nature and is capable of accelerating the growth of dairy starter culture while also producing acid during fermentation.

Table 6. Development of pH in yogurt drink supplemented with *Spirulina* powder

Treatments	Storage	pH
C	Fresh	4.785±0.05
	7 days	4.645±0.04
	15 days	4.560±0.04
T1	Fresh	4.525±0.04
	7 days	4.490±0.014
	15 days	3.695±0.06
T2	Fresh	4.485±0.01
	7 days	4.440±0.014
	15 days	3.415±0.02
T3	Fresh	4.380±0.03
	7 days	4.320±0.014
	15 days	3.360±0.014
T4	Fresh	4.415±0.021
	7 days	4.425±0.04
	15 days	3.510±0.1
T5	Fresh	4.410±0.01
	7 days	4.320±0.01
	15 days	3.180±0.03
T6	Fresh	4.295±0.01
	7 days	4.220±0.01
	15 days	3.195±0.01

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

Color properties

Table 7 shows that as the amount of *Spirulina* powder increased, the L* value, which measures the product's whiteness, decreased significantly (p< 0.05). As the amount of *Spirulina* powder increased, the yogurt drink's greenness increased, which reflected the negative a* value. This may be related to carotene and Chlorophyll (Branger *et al.*, 2003, Habib *et al.*, 2008, Vijayarani *et al.*, 2012, Dolly, 2014 and Sharoba, 2014) the addition of material to yogurt had a significant effect a concentration-dependent-manner lowered the lightness while a* shifted to the positive range with higher values of b* color parameter. Among the most known microalga is *Spirulina* spp. (Arthrosporic) belonging to the cyanobacteria, blue-green colored microalgae which are deemed as intermediate species between plants and bacteria (Parimi *et al.*, 2015).

Table 7. Color properties of yogurt drink supplemented with *Spirulina* powder

		L	A	B
C	Fresh	90.05±0.07	-6.110±0.01414	8.600±0.1414
	7 days	89.20±0.14	-6.110±0.01414	8.200±0.1414
	15 days	88.40±0.14	-6.020±0.02828	8.200±0.2828
T1	Fresh	79.20±0.28	-16.45±0.07	9.950±0.07071
	7 days	78.91±0.007	-16.25±0.07	9.500±0.1414
	15 days	78.11±0.014	-16.01±0.01	9.150±0.07071
T2	Fresh	63.70±0.14	-19.42±0.02828	11.40±0.14
	7 days	63.12±0.028	-19.02±0.02828	11.15±0.07
	15 days	63.11±0.014	-18.99±0.01414	11.00±0.014
T3	Fresh	58.21±0.014	-20.86±0.01000	14.94±0.05
	7 days	57.84±0.05	-20.22±0.02121	14.52±0.028
	15 days	57.64±0.028	-19.91±0.01414	14.13±0.042
T4	Fresh	79.11±0.01	-16.31±0.01	10.02±0.021
	7 days	78.72±0.03	-16.29±0.01	9.810±0.014
	15 days	78.12±0.03	-16.12±0.01	9.515±0.021
T5	Fresh	63.33±0.04	-19.06±0.1	11.82±0.03
	7 days	62.84±0.05	-19.01±0.02	11.28±0.04
	15 days	62.32±0.021	-18.94±0.05	11.12±0.03
T6	Fresh	56.22±0.02828	-20.22±0.02	15.01±0.01
	7 days	55.91±0.01414	-20.03±0.04	14.74±0.06
	15 days	55.62±0.007070	-19.95±0.03	14.11±0.01

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

Phytochemical and antioxidant capacity:

Antioxidant capacity, TP, and TF of yogurt drink enhanced by *Spirulina* powder shown in Table (8) antioxidant, TP, and TF was increased as the level of *Spirulina* powder increased. In agreement with what was found by Vishnu and Sumathi, 2014) who reported that The *Spirulina* platensis powder (SPP) algae has been considered super antioxidants. During storage, antioxidants, total phenolics, and total flavonoids gradually increased for 15 days in both the control group and all treatment groups. Statistics revealed a significant difference between the control and treatment groups.

Diacetyl and Acetoin content

Table 9 shows the diacetyl and acetoin content of a yogurt beverage with *Spirulina* powder added. Diacetyl: The lowest levels of diacetyl were found in control samples. When *Spirulina* powder was added to storage or fresh samples, diacetyl frequently increased. Control and all treatments were periodically increased during the period of cold storage.

Controls and treatments differ significantly, according to statistical analysis. Acetoin followed a similar pattern. These findings follow a similar pattern to El-Sayed *et al.*, (2017).

Table 8. Phytochemical and antioxidant capacity of supplemental drinking yogurt *Spirulina* powder

		Antioxidant (Mg/100)	Total phenolic (Mg/100)	Total flavonoid (Mg/100)
C	Fresh	156.5±0.05	15.34±0.01	108.7±0.02
	7 days	161.1±0.04	17.42±0.06	136.5±0.01
	15 days	169±0.01	20.77±0.04	162.6±0.04
T1	Fresh	169.7±0.021	50.23±0.02	190.5±0.08
	7 days	171.5±0.04	51.86±0.01	232.2±0.03
	15 days	179.7±0.04	62.54±0.04	260.0±0.04
T2	Fresh	174.7±0.06	64.61±0.071	228.7±0.06
	7 days	178.8±0.05	72.53±0.02	254.6±0.06
	15 days	179.7±0.1	76.00±0.01	287.7±0.03
T3	Fresh	204.3±0.06	77.33±0.01	282.6±0.02
	7 days	206.4±0.1	79.23±0.01	299.9±0.18
	15 days	210.4±0.01	80.98±0.01	311.5±0.48
T4	Fresh	195.6±0.03	66.34±0.02121	234.6±0.03
	7 days	216.8±0.014	69.14±0.02828	276.9±0.06
	15 days	220.1±0.02	72.57±0.04	293.4±0.26
T5	Fresh	211.3±0.05	74.67±0.04	306.7±0.04
	7 days	257.0±0.03	79.49±0.06	321.7±0.03
	15 days	273.8±0.04	80.45±0.02	339.4±0.023
T6	Fresh	221.7±0.02	77.56±0.03536	377.5±0.1
	7 days	236.8±0.01	80.36±0.03	397.6±0.02
	15 days	244.6±0.04	87.01±0.04	401.9±0.1

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

Table 9. Diacetyl and Acetoin content of supplemental yogurt drink with *Spirulina* powder

		Diacetyl	Acetoin
C	Fresh	1.068±0.1	1.228±0.01
	7 days	1.226±0.02	1.427±0.01
	15 days	1.419±0.01	1.727±0.01
T1	Fresh	1.360±0.01	1.554±0.01
	7 days	1.548±0.003	1.740±0.004
	15 days	1.648±0.01	1.916±0.02
T2	Fresh	1.649±0.011	1.770±0.01
	7 days	1.778±0.004	1.892±0.001
	15 days	1.801±0.004	1.995±0.01
T3	Fresh	2.022±0.01	2.342±0.001
	7 days	2.129±0.01	2.517±0.02
	15 days	2.458±0.01	2.728±0.01
T4	Fresh	1.471±0.01	1.765±0.003
	7 days	1.683±0.001	1.864±0.001
	15 days	1.858±0.01	2.010±0.01
T5	Fresh	1.757±0.01	1.978±0.
	7 days	1.900±0.004	2.005±0.01
	15 days	2.081±0.1	2.276±0.04
T6	Fresh	2.440±0.03	2.475±0.1
	7 days	2.640±0.01	2.773±0.003
	15 days	2.861±0.01	2.912±0.002

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

α-Tocopherol

α-Tocopherol in yogurt drink supplemented with *Spirulina* powder shown in Table (10) α-Tocopherol was increased as the level of *Spirulina* powder increased, that is because The *Spirulina* platensis powder (SPP) algae has been considered a suitable nutritional additive because it includes vitamin E (Vishnu and Sumathi, 2014) During the time of cold storage, control, and all treatments increased periodically. Statistics revealed a significant difference between the control and treatment groups.

Table 10. α-Tocopherol (µl) in drinking yogurt supplemented with *Spirulina* powder

TREATMENTS	Storage	α-TOCOPHEROL (µl)
C	Fresh	98.53±0.1
	7 days	105.5±0.1
	15 days	110.7±0.2
T1	Fresh	115.6±0.24
	7 days	117.4±0.2
	15 days	125.5±0.2
T2	Fresh	155.7±0.1
	7 days	175.3±0.2
	15 days	185.5±0.2
T3	Fresh	187.7±0.21
	7 days	190.9±0.2
	15 days	193.4±0.1
T4	Fresh	195.7±0.2
	7 days	201.3±0.2
	15 days	203.8±0.2
T5	Fresh	204.5±0.2
	7 days	206.7±0.2
	15 days	215.4±0.3
T6	Fresh	235.6±0.1
	7 days	248.7±0.2
	15 days	265.8±0.1

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

Microbiological analysis:

Table 11's findings demonstrate that there were no yeast or molds in any of the yogurt drink samples. Liveliness of the yogurt drink culture when stored at the recommended temperature of 5°C: Control has less overall lactic acid bacteria overall but can increase *Spirulina* and bifidobacterial counts by 0.5, 1, and 1.5. Up to 15 days of storage, the total lactic acid bacteria and bifidobacterial decreased in the viable count. The yogurt culture's viability decreased as storage time went on, which may have been caused by the period's increased acidity. Laye *et al.,* (1993) reported similar outcomes.

Sensory qualities:

The yogurt drink's sensory qualities are a key factor in determining whether or not consumers will buy it (Table 12). The acceptability of yogurt drinks is significantly influenced by their texture, color, appearance, and flavor. Acceptability of yogurt drinks rose with increasing levels of *Spirulina* enrichment from 0.5 to 1% level, then fell with increasing levels of incorporation up to 1.5%. Yogurt drink acceptance gradually declined throughout storage, lasting 15 days. The ratings for the yogurt beverage that had a 1% *Spirulina* enrichment were higher than the ratings for the control. This might be explained by how *Spirulina* works. Comparing the experimental product to the control, the experimental product received higher ratings for flavor, texture, color, and appearance.

Table 11. Microbiological analysis drinking yogurt supplemented with *Spirulina* powder

		LAB (10^8)	B.longum (10^7)	molds	Yeasts
C	Fresh	6	-	-	-
	7 days	8	-	-	-
	15 days	6	-	-	-
T1	Fresh	20	-	-	-
	7 days	25	-	-	-
	15 days	22	-	-	-
T2	Fresh	28	-	-	-
	7 days	30	-	-	-
	15 days	28	-	-	-
T3	Fresh	33	-	-	-
	7 days	36	-	-	-
	15 days	34	-	-	-
T4	Fresh	36	40	-	-
	7 days	45	48	-	-
	15 days	29	45	-	-
T5	Fresh	61	80	-	-
	7 days	70	84	-	-
	15 days	66	81	-	-
T6	Fresh	81	104	-	-
	7 days	84	112	-	-
	15 days	82	110	-	-

C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina.

Table 12. Sensory characteristics of yogurt drink. enriched with *Spirulina* powder

		Flavor 20	Texture 10	Color 10	Acceptability 40
C	Fresh	18.40±0.7	9.300±0.7	8.100±0.7	39.50±0.7
	7 days	18.30±0.7	9.500±0.7	7.900±0.7	39.50±0.7
	15 days	15.50±0.7	6.500±0.7	7.500±0.7	35.50±0.7
T1	Fresh	19.00 ±0.7	9.500±0.7	9.500±0.7	39.93±0.7
	7 days	19.20±0.7	9.500±0.7	9.200±0.7	39.50±0.7
	15 days	12.00±0.01	7.500±0.7	7.500±0.7	34.07±.0.7
T2	Fresh	19.50±0.7	9.500±0.7	9.800±0.7	39.95±0.7
	7 days	17.50±0.7	9.500±0.7	9.52±0.7	36.50±0.7
	15 days	11.50±0.7	7.500±0.7	7.500±0.7	36.50±0.7
T3	Fresh	17.50±0.7	7.500±0.7	7.500±0.7	36.50±0.7
	7 days	16.50±0.7	7.500±0.7	7.500±0.7	34.50±0.7
	15 days	9.500±0.7	7.500±0.7	7.500±0.7	27.50±0.7
T4	Fresh	19.50±0.7	9.500±0.7	9.400±0.7	39.80±0.7
	7 days	17.50±0.7	9500±0.7	9.500±0.7	37.50±0.7
	15 days	17.50±0.7	7.500±0.7	8.500±0.7	27.50±0.7
T5	Fresh	19.70±0.7	9.57±0.7	9.850±0.7	39.94±0.7
	7 days	16.50±0.7	9.550±0.7	9.500±0.7	29.50±0.7
	15 days	14.50±0.7	7.500±0.7	6.500±0.7	29.50±0.7
T6	Fresh	17.50±0.7	7.500±0.7	7.500±0.7	31.50±0.7
	7 days	15.50±0.7	5.500±0.7	6.500±0.7	28.50±0.7
	15 days	11.50±0.7	5.500±0.7	6.500±0.7	26.50±0.7

Values are expressed as mean± standard deviation (N=3). a (significantly different from the control group at P < 0.05, C; control, T1; drinking yogurt culture with adding 0.5% Spirulina powder and, T2; drinking yogurt culture with adding 1% Spirulina powder T3; drinking yogurt culture with adding 1.5% Spirulina powder T4; drinking yogurt culture and bifidobacterial culture with adding 0.5% Spirulina powder, T5; drinking yogurt culture and bifidobacterial culture with adding 1% Spirulina powder, T6; drinking yogurt culture and bifidobacterial culture with adding 1.5% Spirulina

CONCLUSION

Overall, our findings demonstrated that adding *Spirulina* to yogurt drinks 0.5, 1, and 1.5% increased its nutritional value without affecting consumer acceptance. This study outlined the qualities that still require improvement and illustrated the value of early-stage consumer acceptance studies in guiding the creation of new products**.**

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تعزيز القيمة الغذائية والتركيب الكيميائي لمشروب الزبادي الوظيفي بإضافة عسل النحل ومسحوق سبيرولينا

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[3]قسم وقاية النبات – كلية الزراعة – جامعة المنيا

الملخص

طحلب الاسبيرولينا غني في محتواه من البروتين والكاروتينات والفيتامينات. في هذه الدراسة تم إضافة مسحوق الاسبيرولينا المجفف الي مشروب الزبادي بثلاث تركيزات 0.5 – 1 – 1.5 % وتم تقسيم هذه التجربة الي جزئين الجزء الأول استخدام البادئ الزبادي التقليدي مع هذه النسب والجزء الثاني عبارة عن مشروب زبادي مثل الجزء الأول ولكن مضاف إليه بيفيدو بكتريا وأظهرت الدراسة أن الجوامد الكلية والتوكوفيرول والرماد يزداد بزيادة تركيز الاسبيرولينا أما بالنسبة لـ pH يقل بزيادة الاسبيرولينا وأثناء التخزين ايضاً. أما بالنسبة لمواد النكهة مثل الداي اسيتيل والاسيتوين فان الطحلب زيادته تؤدي الي زيادة تكون هذه المواد وهذا يظهر ايضاً بالنسبة لمضادات الأكسدة والفينولات والفلافونيدات اما من ناحية الاختبارات الحسية فان اضافة الاسبيرولينا نسبة 1% وجد قبولا عاليا تقريبا مثل الكنترول. أما بالنسبة للعد البكتيري فان عدد البكتريا عموما يزداد بزيادة تركيز الاسبيرولينا ولكن اثناء التخزين يزداد العدد الكلي حتى اليوم السابع ثم يقل بعد ذلك.