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The Effects of Adding *Chlorella Vulgaris* on Sponge Cake Quality and Nutritional Value

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ABSTRACT

As one in pursuing novel food ingredients that transcend seasonal limitations and boast an abundance of bioactive compounds, scientists have turned their attention to microalgae. This study search into the enrichment of cakes with microalgae powder derived from *Chlorella vulgaris* (C.V). Fortification of 0.5%, 1.0%, and 2.0% C.V were employed. A comprehensive evaluation of the cakes' physical, chemical, quality characteristics, and sensory attributes was conducted. Additionally, the cakes were stored at 25°C for 21 days, with storage assessments performed at intervals of 0, 1, 2, and 3 weeks. The study included extracting the active substances from *Chlorella vulgaris* (C.V) using three solvents: 50% ethanol, 95% ethanol, and water. Descriptive analysis of the extracts revealed the presence of various compounds such as phenols, alkaloids, flavonoids, glycosides, and saponins. The results indicated that the cake fortified with C.V exhibited excellent stability against oxidation compared to the control. The addition of C.V to the cake resulted in a noticeable reduction in microbial load compared to the control. However, sensory evaluation revealed that the cakes containing C.V exhibited slightly lower sensory quality compared to the control. Nevertheless, the findings of this study unequivocally demonstrate the effectiveness of C.V in minimizing microbial contamination and extending the shelf life of cakes due to their natural antioxidant properties also enhances their nutritional quality. These findings highlight the potential of C.V as a natural ingredient with antimicrobial properties and antioxidant capacity, which can effectively improve the microbial safety and shelf stability of food products like cakes.

1. Introduction

Microalgae are a diverse group of single-celled organisms that are found in both freshwater and marine environments and have a wide range of potential applications. They are a rich source of essential fatty acids, pigments, antioxidants, and other bioactive compounds that have a variety of health benefits. As a result, microalgae are being investigated for use in a variety of industries, including food and beverage, pharmaceuticals, and cosmetics [1].

Chlorella vulgaris is a specific species of microalgae categorized under the *Chlorococcales* order within the *Oocytaceae* family and the genus Chlorella. it is a microalgae species rich in proteins, carbohydrates, lipids, and various vitamins and pigments. is a unicellular organism characterized by a spherical shape and varying sizes ranging from 2 to 10 microns [2]. *Chlorella vulgaris* is renowned for its diverse range of functional macronutrients and micronutrients, encompassing omega-3 polyunsaturated fatty acids, polysaccharides, minerals, chlorophyll, vitamin C, β-carotenes, and B vitamins (B1, B2, B6, and B12) [3].

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Chlorophyll is a dominant pigment in *C. vulgaris*, making up approximately 1 to 2% of the dry weight of its biomass. *C. vulgaris* contains various primary pigments, including β-carotenes, astaxanthin, canthaxanthin, lutein, chlorophyll a and b, pheophytin a and b, and violaxanthin. Additionally, the presence of several polyphenols, such as luteolin, can also be observed in *C. vulgaris* [4]. It has gained popularity as a dietary supplement and has been associated with numerous health advantages such as improving hyperlipidemia, hyperglycemia, safeguarding against oxidative stress, cancer, and chronic obstructive pulmonary disease [5].

Chlorella vulgaris has been studied for its antioxidant activity and potential applications. Research has shown that *Chlorella vulgaris* has antioxidant and anticancer activities [6,7]. The antioxidant activity of *Chlorella vulgaris* has been evaluated using various methods, including the DPPH and FRAP assays. The results of these studies have shown that *Chlorella vulgaris* has high antioxidant activity, which is attributed to its unique and diverse composition of functional macro- and micro-nutrients [8]. *Chlorella vulgaris* has also been studied for its potential applications in the food, pharmaceutical, and cosmetic industries. For example, *Chlorella vulgaris* cream has been shown to have a high antioxidant activity and can be used as an anti-aging cream [9]. Its nutritional composition and functional qualities make it a valuable ingredient for dietary supplements and functional foods. With its natural emulsifying, stabilizing, and thickening properties, *Chlorella vulgaris* enhances food texture and stability [10]. Incorporating it into food formulations can improve nutritional content and offer potential health benefits, making it an increasingly important component in the food industry.

Cereal-based products are widely consumed regularly by people all over the world. Cakes and other bakery products are popular food choices and are often rich in carbohydrates, calories, and fats while lacking in fiber, vitamins, and minerals. However, there is a growing demand for healthier food options in the food industry. As a response, efforts have been made to develop food products, particularly in the realm of bread products like cakes, that are high in dietary fibers and antioxidants. These additions aim to enhance the nutritional value of these baked goods and promote healthier consumption choices [11].

To enhance the quality of cakes and address the deficiency of functional and nutritional components, it is important to develop fortified cake products that incorporate a substantial amount of plant-based ingredients while maintaining satisfactory sensory attributes. In line with this objective, the present study aims to evaluate the impact of substituting wheat flour with *Chlorella vulgaris* powder on the functional, nutritional, and sensory properties of sponge cakes.

2. Materials and Methods

2.1. Raw Material

Chlorella vulgaris, a freshwater green algae belonging to the Chlorophyta phylum, was sourced from the Algal Biotechnology Unit at the National Research Centre in Giza, Egypt.

2.2. Chemicals, Solvents, reagents, and media

Aluminum chloride ($AlCl_3$), sodium carbonate, ethanol, hexane, hydrochloric acid, boric acid, sodium hydroxide, glacial acetic acid, thiobarbituric acid, soluble starch, sodium thiosulphate, sulphuric acid, and total count agar was sourced from El-Gomhoria Company (Egypt). Methanol was purchased from Aldrich Co. Folin–Ciocalteu reagents were obtained from Sigma Chemical Co. All other solvents and chemicals used in the study were of analytical grade.

2.3. Methods

2.3.1. Preparation of the Extract

C. vulgaris crude extracts were made with ethanol 95, ethanol 50%, and water. Ten grams of *C. vulgaris* biomass and 100 mL of each solvent were combined in a conical flask and placed on a shaking water bath at 25°C with a shaking rate of 200 rpm per minute for 3 hours. The mixture was centrifuged using 10000 rpm centrifugation. Re-maceration was carried out three times until the colour of the mixture vanished. The filtrate was extensively dried in a hot air oven at 40°C.

2.3.2. Cakes Preparation

To prepare the cake batter, 200 grams of flour, 120 grams of sugar, 100 milliliters of skimmed milk, 80 grams of fresh whole eggs, 100 grams of butter, 8 grams of baking powder, and 4 grams of vanilla were mixed in a Kitchen-Aid Professional mixer and blended thoroughly for 10 minutes at a specified speed. The resulting batter was poured into metallic pans measuring 120 millimeters in diameter and 45 millimeters in height, which had been previously greased with butter. The pans were then placed in an electric oven preheated to 200°C and baked for 30 minutes [12].

To investigate the effects of *Chlorella vulgaris* powder on the cake, different substitution levels of wheat flour were tested: 0% (used as a control), 0.5%, 1%, and 2%. After baking the cakes were allowed to cool for 30 minutes, they were packed and assessed for their chemical, physical, and sensory characteristics. Subsequently, the cakes were stored in polyethylene bags at room temperature for approximately 21 days to enable further analysis.

2.3.3 Characteristics of cakes

After baking the control and treatments were left for three hours at room temperature then, the cakes were weighed. The Alfalfa seeds replacement method, as outlined in the [13,14] reference, was used to determine the volume of the cakes. The specific volume was calculated by dividing the volume of each cake by its corresponding weight.

2.3.4. Chemical composition of cakes:

Moisture, protein, ash and ether extract contents were determined according to [15].

2.3.5. Determination of total phenols, flavonoids, and antioxidant activity of cakes

The measurement of the total phenolic content (TPC) in the sponge cakes was conducted based on a previously established method [16]. Similarly, the determination of the total flavonoid content (TFC) was carried out following a method described in a previous study [17]. The antioxidant activity

of the sponge cakes was assessed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging method as outlined by [18,19].

2.3.6. Determination of oxidative stability

One hundred grams of Roughly ground cakes were weighed then, transferred into a sealed flask, followed by the addition of 200 ml of *n*-hexane as a solvent. The flask was vigorously shaken for 1 hour, after which the mixture was filtered to separate the extracted fat. The solvent was subsequently evaporated from the extracted fat by using a rotary evaporator at 50ºC. The extracted fat obtained from this process was then utilized for several analyses, which will be detailed in the forthcoming sections. Peroxide value was determined according to [20]. The Thiobarbituric Acid (TBA) values of the cake's extracts were determined using the [21].

2.3.7. Microbiological Examination

The plate count of aerobic bacteria in the cakes was conducted according to the guidelines outlined by [22].

2.3.8. Sensory Evaluation:

An assessment of the cake's quality attributes through sensory evaluation was conducted after allowing it to cool to room temperature for 2 hours. Eleven trained panelists, consisting of graduate students and staff members from the Department of Food Science and Technology at Fayoum University, participated in the evaluation. Each panelist was randomly assigned cakes to assess. The panelists were instructed to evaluate each cake based on taste, odor, texture, appearance, color, mouthfeel, and overall acceptability. A 10-point scale was employed for rating, with 10 indicating "excellent" and 1 representing "extremely unsatisfactory," following the guidelines provided by the [14].

2.3.9. Statistical analyses

The experimental design employed in this study was a randomized complete block design, consisting of four treatments and ten replicates, as described by [23]**.** The collected data was subjected to analysis of variance (ANOVA) following the general linear model approach. For statistical analysis, the Info Stat computer software program (version 2012) was utilized. To determine significant differences between means at a 95% confidence level, Duncan's multiple range test was applied.

3. Results and Discussion

3.1. Characteristics of Cake

The alterations in cake properties upon the incorporation of *Chlorella vulgaris* (C.V) powder are presented in Table 1, Both the weight and volume of the cake exhibited variations. An evident increase in cake size occurred with escalating levels of C.V powder. The average volume of the control sample was 326 ml, while it increased to 343 ml, 368 ml, and 394 ml for 0.5%, 1%, and 2% C.V, respectively. Previous studies [24] have suggested that an optimal cake batter should possess appropriate viscosity to prevent the premature rise and escape of air bubbles during the initial heating stages.

Table 1: The physical attributes of cakes produced by substituting *Chlorella vulgaris* powder for cake.

Character	Substitution level			
	0% Algae	0.5%Algae	1.0%Algae	2.0%Algae
Weight	136.86	137.34	139.81	143.21
Volume	326	343	368	394
Specific Volume	$2.38^{c}\pm0.10$	$2.50^{bc}\pm0.08$	$2.63^{ab}\pm0.12$	$2.75^{a}\pm0.10$

Means in row designated with the same letter are not significantly different at (P > 0.05)

The timing of the cake preparation process is crucial to allow sufficient expansion of air bubbles due to the presence of carbon dioxide gas and water vapor before full baking. This careful timing ensures a lighter texture and well-defined structure in the resulting cake. In our investigation, the volume of cake mixture with 0.5%, 1%, and 2% C.V exceeded both the control volume and their respective specific volumes as shown in Figure 1. The addition of C.V at levels 1 and 2% significantly increases the specific volume of cake in comparison with the control, this may be due to the ability of *Chlorella vulgaris* as an emulsifying agent, as explained by [25], who applied chlorella to emulsions and found that it improved the stability of the emulsion. *Chlorella vulgaris* also has viscoelastic qualities which makes it work as functions as a natural emulsifier, stabilizer, and thickening agent, hence improving product consistency and stability [10,26,27].

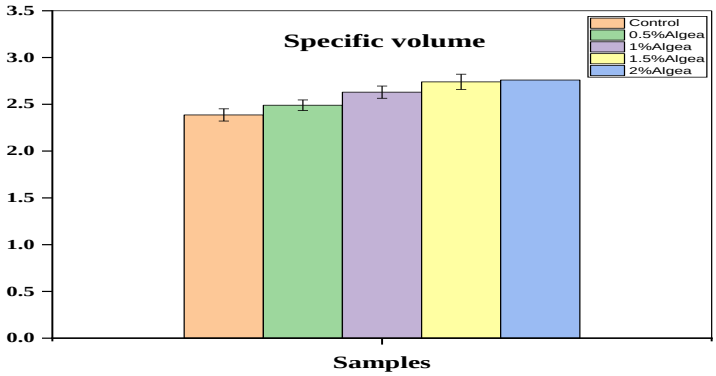

Fig.1 Specific volume of cake samples as affected by C.V addition.

3.2. The proximate analysis of cakes treated with *Chlorella vulgaris* powder.

Distinct proximate compositions were observed among the cakes, as detailed in Table 2. The total ash and protein contents exhibited a descending order of 2% > 1% > 0.5% > Control, indicating clear and significant differences between the treatments. This finding aligns with previous studies by [28], who incorporated chlorella powder into pasta. In contrast, moisture and fat contents did not exhibit clear significant differences among the treatments (0.5%, 1%, and 2%) and control. Table (2) shows that the ash content of cake increases with increasing C.V addition. The elevated ash and protein content in the samples may be attributed to the higher *C. vulgaris* content of these substances compared to flour.

Table (2) shows the content of control, 0.5%, 1%, and 2% C.V from total phenols and total flavonoids. The trend in phenols and flavonoids exhibited a positive correlation with the incorporation of C.V powder into the cake formulation. Specifically, the result showed that total phenols and total flavonoids increase with the increasing addition of C.V powder to the cake. The formulation with 2% C.V powder demonstrated the highest Total Phenolic Content (TPC) compared to control cakes.

Table 2: The proximate chemical composition of cakes is prepared by replacing cake flour with *Chlorella vulgaris* powder.

Treatments	Chemical Composition%				T. Phenols (mg/g)	T. Flavonoids (mg/g)	Antioxidant activity Inhibition %
	Moisture	Protein	Ether extract	Ash			
Control	$19.87^{a}\pm0.85$	$13.50^{d}\pm0.20$	$24.33^{a}\pm0.25$	$1.13^{d}\pm0.13$	$4.98^{d}\pm0.16$	$2.29^{d}\pm0.10$	$6.28^{d}\pm0.40$
0.5%Algea	$20.07^{a}\pm0.45$	$14.25^{c}\pm0.07$	$23.35^{b}\pm0.07$	$1.33^{c}\pm0.15$	$8.76^{c}\pm0.09$	$2.53^{c}\pm0.10$	$18.73^{c}\pm1.12$
1.0%Algea	$20.20^{a}\pm0.53$	$14.70^{b}\pm0.14$	$23.60^{ab}\pm0.14$	$1.67^{b}\pm0.14$	$9.58^{b}\pm0.06$	$2.82^{b}\pm0.10$	$21.00^{b}\pm0.89$
2.0%Algea	$20.40^{a}\pm0.25$	$15.30^{a}\pm0.14$	$24.10^{a}\pm0.14$	$1.95^{a}\pm0.14$	$10.56^{a}\pm0.10$	$2.92^{a}\pm0.02$	$23.08^{a}\pm0.64$

Means in each column identified with the same letter indicate no significant differences ($p > 0.05$).

3.3. Antioxidant activities in cake

Table (2) percentage of inhibition of DPPH• free radicals for various cake formulations enriched with *Chlorella vulgaris* (C.V) powder. The addition of C.V powder into the cakes led to a substantial enhancement in antioxidant activity, surpassing that of control cakes without C.V powder (Control < 0.5% < 1% < 2%). The average inhibition percent of DPPH• free radical capacity of the control sample was 6.28%, while it increased to 18.73%, 21%, and 23.8% for 0.5%, 1%, and 2% C.V, respectively.

This association is primarily attributed to the significant presence of phytochemicals, particularly phenolic compounds, in C.V, which exhibit robust antioxidant activity [7]. Therefore, the incorporation of C.V into cakes not only improves their antioxidant potential but also enhances their nutritional quality and stability against oxidation.

3.4. Oxidative stability of Cake

The oxidation of lipids represents a prevalent process in cakes with elevated lipid contents. The hydrolytic degradation of lipids is a primary precursor to oxidative instability in bakery products, especially under high baking temperatures. Various lipid peroxidation compounds, including aliphatic aldehydes and 1-octanol, have been reported to be detected in sponge cakes during the baking process [29].

Table 3 presents the levels of lipid oxidation products, expressed as Peroxide value and TBARS, in *Chlorella vulgaris* (C.V) supplemented sponge cakes. The addition of C.V at concentrations 1% and 2% resulted in a significant reduction in peroxide values in sponge cakes compared to the control sponge cake, while there is no significant difference between concentration 0.5% and control. Notably, even a concentration of 2% C.V demonstrated a capacity to decrease lipid oxidation in sponge cake. Various studies have highlighted the antioxidant activity of C.V, as indicated by assays such as Ferric ion reducing antioxidant power (FRAP), ABTS, and DPPH [7,30]. Prior research has also demonstrated that C.V at concentrations of 0.05% to 0.1% w/w, containing phenolic compounds, effectively inhibits lipid oxidation in rainbow trout (Oncorhynchus mykiss) fillets during refrigerated storage [8].

Table 3: Changes in Thiobarbituric acid value and Peroxide value in cakes during storage period (Room Temperature).

Parameters	Storage time (Weeks)	Treatments			
		Control	0.5%C.V	1.0%C.V	2.0%C.V
PV (m.eq. of O_2 /kg of oil)	0	1.02±0.29	1.97±0.23	1.96±0.17	1.36±0.21
	1	3.15±0.19	3.11±0.09	2.49±0.13	1.92±0.10
	2	6.86±0.36	4.92±0.27	4.08±0.23	2.39±0.30
	3	13.65±0.32	5.73±0.22	4.67±0.18	3.06±0.24
	Mean	6.17[a]	3.93[ab]	3.30[b]	2.19[b]
TBARS (MDA µg/g of oil)	0	0.41±0.18	0.36±0.11	0.39±0.20	0.48±0.17
	1	1.47±0.15	1.26±0.22	1.27±0.11	0.96±0.19
	2	1.95±0.24	1.65±0.18	1.38±0.26	1.35±0.28
	3	2.94±0.10	1.8±0.23	1.69±0.21	1.39±0.11
	Mean	1.69[a]	1.27[ab]	1.18[b]	1.05[b]

Means in each row identified with the same letter indicate no significant differences ($p > 0.05$).

This investigation suggests that a higher concentration of C.V may yield a more pronounced inhibition effect on PV compared to a control. The results indicate a concentration-dependent variation in the antioxidant activity of C.V. Supporting this, a positive correlation between the phenolic compound content in *Chlorella vulgaris* and the inhibition of lipid oxidation in fish burgers has been reported [31].

The formation of lipid peroxidation gives rise to a secondary product, malondialdehyde (MDA). As indicated in Table 3, the TBA (thiobarbituric acid) values for the reformulated cake samples (0.5%, 1.0%, and 2.0%) were consistently lower than those of the control sample. Notably, *Chlorella vulgaris* (C.V) significantly reduced TBA values compared to control values throughout the storage period at concentrations of 1.0% and 2.0% same trende was observed by [32]. For cake samples, a TBA value below 0.6 mg/kg MDA is considered non-rancid, while values falling within the range of 0.65 to 1.44 mg/kg MDA are considered rancid but still acceptable. Values exceeding 1.5 mg/kg of the sample are deemed rancid and unacceptable [12].

According to the [12] scale on the seventh day of storage, the control sample exhibited signs of spoilage but remained acceptable. In contrast, the samples treated with 1% and 2% *Chlorella vulgaris* remained acceptable until day 14, meanwhile, 2% concentration remained acceptable until 21 days. The control sample displayed the highest TBA value compared to the other reformulated cake samples, suggesting that the addition of C.V as an antioxidant component can extend the shelf life of cakes by inhibiting oxidation reactions.

3.5. The alteration in the total bacterial count of cake samples during a three-week storage period.

Figure 2 illustrates the total bacterial counts of various cake samples. The results indicate a significant decrease in the number of colonies, specifically in samples treated with 0.5%, 1%, and 2% of *Chlorella vulgaris* powder. This reduction in bacterial colonies could be attributed to the antimicrobial effects associated with the presence of algal biomass.

This finding is consistent with studies conducted by [33] then demonstrated that *Chlorella vulgaris* exhibits antimicrobial activity.

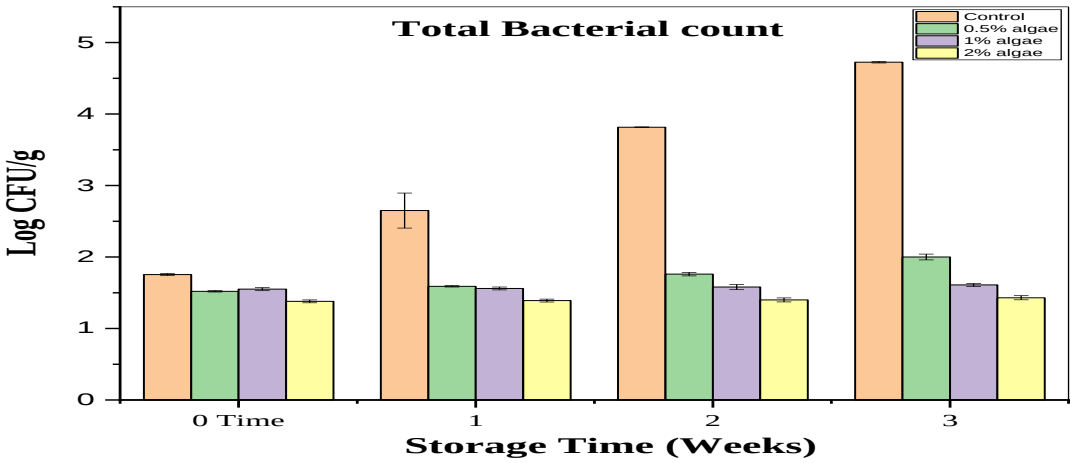

Fig.2 Change in the total bacterial count of cake samples during a three-week storage period

3.7. The Sensory Evaluation of Cakes:

Cake samples underwent organoleptic evaluation, and the results were subjected to statistical analysis, as depicted in Table 4. The treatment with the highest average score is considered superior in terms of determined quality characteristics. In summary, the statistical analysis of sensory characteristics indicated that the samples treated with *Chlorella vulgaris* powder at concentrations of 0.5%, 1%, and 2% were significantly lower than the control in terms of sensory attributes.

Table 4: Sensory evaluation of experimental Cake samples:

Cake Samples	Control	0.5%C.V	1.0%C.V	2.0%C.V
Taste	$9.00^{a}\pm0.82$	$8.20^{ab}\pm0.92$	$8.30^{ab}\pm0.48$	$7.40^{b}\pm1.51$
Odor	$9.00^{a}\pm1.15$	$8.80^{a}\pm0.63$	$8.40^{ab}\pm0.84$	$7.70^{b}\pm1.77$
Texture	$9.40^{a}\pm0.52$	$8.90^{b}\pm0.74$	$8.80^{b}\pm0.79$	$8.40^{c}\pm0.84$
Appearance	$9.60^{a}\pm0.52$	$8.60^{b}\pm0.70$	$7.70^{b}\pm1.49$	$6.80^{c}\pm1.93$
Color	$9.70^{a}\pm0.48$	$7.80^{b}\pm1.03$	$6.90^{bc}\pm1.73$	$6.00^{c}\pm1.70$
Mouth feel	$9.20^{a}\pm0.79$	$8.30^{b}\pm0.67$	$8.30^{b}\pm0.95$	$7.90^{b}\pm1.10$
Overall	$9.40^{a}\pm0.70$	$8.30^{b}\pm0.82$	$7.40^{bc}\pm1.35$	$7.10^{c}\pm1.66$

Means in each row identified with the same letter indicate no significant differences ($p > 0.05$).

4. Conclusions

The incorporation of *C. vulgaris* biomass as a natural ingredient has resulted in cakes with an appealing and innovative appearance, along with enhanced textural characteristics. The comprehensive analysis covering chemical and microbial aspects suggests that *Chlorella vulgaris* (C.V) can serve as a promising substitute for synthetic antioxidants and preservatives. The heightened efficacy of the chlorella in slowing down lipid oxidation is likely attributed to its phenolic content and the stability of this natural antioxidant during the baking process. These findings have significant implications for industries seeking to leverage the nutritional benefits of *Chlorella vulgaris* as a supplement to cereal flour. C.V proves to be a valuable resource to produce highly nutritious cakes, offering functional advantages.

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Author Contributions

All authors contributed to this work. B. Abdel-moatamed prepared the samples and completed the experimental measurements. Both A. Elfakharany and N. Elneary shared writing and followed the performance of the experiments. M. Shaban helped the first author in complete the sample preparation and analysis measurments. M. Roby with N. Elneary completed the paper writing, analyzing the data, and validation. M. Roby followed the revision and submission of the manuscript for publication.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References:

[1] M. Rizwan, G. Mujtaba, S.A. Memon, K. Lee, N. Rashid, Exploring the potential of microalgae for new biotechnology applications and beyond: A review, Renew. Sustain. Energy Rev., 92 (2018) 394–404.
[2] J.A. Coronado-Reyes, J.A. Salazar-Torres, B. Juárez-Campos, J.C. González-Hernández, Chlorella vulgaris, a microalgae important to be used in Biotechnology: a review, Food Sci. Technol., 42 (2022).
[3] D. Widyaningrum, A.D. Prianto, Chlorella as a Source of Functional Food Ingredients: Short review, in: IOP Conf. Ser. Earth Environ. Sci., IOP Publishing Ltd, 2021.
[4] R. Neenu, S. Tiwari, H. Jethani, V.S. Chauhan, The commercial microalgae-based foods, in: Handb. Food Feed from Microalgae Prod. Appl. Regul. Sustain., 2023.
[5] T. Maurício, D. Couto, D. Lopes, T. Conde, R. Pais, J. Batista, T. Melo, M. Pinho, A.S.P. Moreira, M. Trovão, A. Barros, H. Cardoso, J. Silva, P. Domingues, M.R. Domingues, Differences and Similarities in Lipid Composition, Nutritional Value, and Bioactive Potential of Four Edible Chlorella vulgaris Strains, Foods, 12 (2023).
[6] R.A. Hamouda, A.A. El Latif, E.M. Elkaw, A.S. Alotaibi, A.M. Alenzi, H.A. Hamza, Assessment of Antioxidant and Anticancer Activities of Microgreen Alga Chlorella vulgaris and Its Blend with Different Vitamins, Molecules, 27 (2022).
[7] K. Mtaki, M.S. Kyewalyanga, M.S.P. Mtolera, Assessment of antioxidant contents and free radical-scavenging capacity of chlorella vulgaris cultivated in low cost media, Appl. Sci., 10 (2020) 1–11.
[8] M.B.T. Takyar, S.H. Khajavi, R. Safari, Evaluation of antioxidant properties of Chlorella vulgaris and Spirulina platensis and their application in order to extend the shelf life of rainbow trout (Oncorhynchus mykiss) fillets during refrigerated storage, LWT, 100 (2019) 244–249.
[9] T.W. Putri, N.F. Nursida, I. Raya, Antioxidant Activity of Chlorella vulgaris Used As an Antioxidant Cream, in: J. Phys. Conf. Ser., 2021.
[10] P. Y., D. B., J. N., B. F., A. Sahebkar, Chlorella vulgaris: a multifunctional dietary supplement with diverse medicinal properties, Curr. Pharm. Des., 22 (2016) 164–173.
[11] S.P. Bangar, A.K. Siroha, Functional Cereals and Cereal Foods: Properties, Functionality and Applications, 2022.
[12] A.A. Hafez, Physico-Chemical and Sensory Properties of Cakes Supplemented with Different Concentration of Marjoram, Aust. J. Basic Appl. Sci., 6 (2012) 463–470.
[13] A. Hosseini, F. Pazhouhandeh, Production of enriched cakes by apple pulp and peel powder and evaluation of chemical, functional and textural properties, Vitae, 30 (2023).
[14] AACC, Cereal Laboratory Methods, Method 46-09 American Association of Cereal Chemists, 7th Ed, (Am. Assoc. Cereal Chem., St. Paul, MN), 1983.
[15] AOAC, Official methods of Analysis, 16th ed Association of Official Analytical Chemistry (Virginia Arlington, USA), 2005.
[16] X. Feng, G. Sun, Z. Fang, Effect of Hempseed Cake (Cannabis sativa L) Incorporation on the Physicochemical and Antioxidant Properties of Reconstructed Potato Chips, Foods, 11 (2022).

[17] S. P., S. S., B. Singh, Evaluation and quality assessment of defatted microalgae meal of Chlorella as an alternative food ingredient in cookies, Nutr. Food Sci., 49 (2018) 221–231.
[18] L. Msaddak, R. Siala, N. Fakhfakh, M.A. Ayadi, M. Nasri, N. Zouari, Cladodes from prickly pear as a functional ingredient: Effect on fat retention, oxidative stability, nutritional and sensory properties of cookies, Int. J. Food Sci. Nutr., 66 (2015) 851–857.
[19] M.H.H. Roby, M.A. Sarhan, K.A.H. Selim, K.I. Khalel, Evaluation of antioxidant activity, total phenols and phenolic compounds in thyme (Thymus vulgaris L), sage (Salvia officinalis L), and marjoram (Origanum majorana L) extracts, Ind. Crops Prod., 43 (2013) 827–831.
[20] M. Kozłowska, A. Żbikowska, A. Szpicer, A. Półtorak, Oxidative stability of lipid fractions of sponge-fat cakes after green tea extracts application, J. Food Sci. Technol., 56 (2019).
[21] AOCS, Official Methods and Recommended Practices of the American Oil Chemist, Soc. Champaign, IL., (2016).
[22] I.S. Organization, ISO 7218 Microbiology of food and animal feeding stuffs - General requirements and guidance for microbiological examinations, Microbiol. Des Aliment. - Exig. Générales Recomm., (2007).
[23] A.A. Gomez, A.A. GOMEZ, STATISTICAL PROCEDURES FOR AGRICULTURAL RESEARCH Second Edition, A Wiley-Intersclence Publ. JOHN WILEY SONS, New York, 6 (1984).
[24] T.M. Ali, M. Shaikh, T. Mehfooz, A. Hasnain, Cakes and Muffins In Cereal-Based Food Products, Cham: Springer International Publishing, 2023.
[25] L. Gouveia, A. Raymundo, A.P. Batista, I. Sousa, J. Empis, Chlorella vulgaris and Haematococcus pluvialis biomass as colouring and antioxidant in food emulsions, Eur. Food Res. Technol., 222 (2006) 362–367.
[26] G. L., B. A.P., S. I., A. Raymundo, N.M. Bandarra, Food Chemistry Research Developments “Microalgae in novel food products,” Ed, Nova Science Publishers, New York, NY., 2008.
[27] A.P. Batista, L. Gouveia, N.M. Bandarra, J.M. Franco, A. Raymundo, Comparison of microalgal biomass profiles as novel functional ingredient for food products, Algal Res., 2 (2013).
[28] Ḿonica Fradique, A.P. Batista, M.C. Nunes, L. Gouveia, N.M. Bandarra, A. Raymundo, Incorporation of Chlorella vulgaris and Spirulina maxima biomass in pasta products Part 1: Preparation and evaluation, J. Sci. Food Agric., 90 (2010) 1656–1664.
[29] R. Zamora, F.J. Hidalgo, The triple defensive barrier of phenolic compounds against the lipid oxidation-induced damage in food products, Trends Food Sci. Technol., 54 (2016) 165–174.
[30] I.K.R. Tiong, T. Nagappan, M.E.A. Wahid, T.S.T. Muhammad, T. Tatsuki, W.H. Satyantini, G. Mahasri, P. Sorgeloos, Y.Y. Sung, Antioxidant capacity of five microalgae species and their effect on heat shock protein 70 expression in the brine shrimp Artemia, Aquac. Reports, 18 (2020).
[31] A. Ben Atitallah, F. Hentati, M. Dammak, B. Hadrich, I. Fendri, M.A. Ayadi, P. Michaud, S. Abdelkafi, M. Barkallah, Effect of microalgae incorporation on quality characteristics and functional and antioxidant capacities of ready-to-eat fish burgers made from common carp (Cyprinus carpio), Appl. Sci., 9 (2019).
[32] B.R. Abdel-Moatamed, A.-E.M.A. El-Fakhrany, N.A.A. Elneairy, M.M. Shaban, M.H.H. Roby, The Impact of Chlorella vulgaris Fortification on the Nutritional Composition and Quality Characteristics of Beef Burgers, Foods, 13 (2024).
[33] S. Syed, A. Arasu, I. Ponnuswamy, The uses of Chlorella Vulgaris as antimicrobial agent and as a diet: The presence of bio-active compounds which caters the vitamins, minerals in general, Int. J. Bio-Science Bio-Technology, 7 (2015) 185–190.