

Research Article

Improvement of Nutritional and Quality of Lasagna Supplemented with *Scenedesmus obliquus*Farouk K. El-Baz^{*1}, Azza A. Omran², Ahmed M.S. Hussein³, Eman A. Ibrahim¹¹Department of Plant Biochemistry, National Research Centre (NRC), 33 El Bohouth st. (former El Tahrir st.), Dokki, Giza, Egypt.²Department of Crops Technology Research, Food Technology Research Institute, Agricultural Research Centre (ARC), Giza, Egypt.³Department of Food Technology, National Research Centre (NRC), 33 El Bohouth st. (former El Tahrir st.), Dokki, Giza, Egypt.*Corresponding author's E-mail: fa_elbaz@hotmail.com

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ABSTRACT

The present work aimed to investigate the effect of mixing microalgae *Scenedesmus obliquus* with wheat flour (72% extraction rate) on enhancing the nutritional characteristics, phytochemical compounds and antioxidant activity of lasagna pasta. Chemical analysis showed that, *S. obliquus* powder had high contents of protein, crude fiber, fats and ash (52.43, 4.53, 2.02 and 6.13%, respectively). Also, it contained high amounts of phytochemical compounds, consequently characterized with high antioxidant activity. Furthermore, it was safe as an edible source, where its lethal toxicity was 10 g/kg mice body weight corresponding to 78 g/70 kg human body weight. Results also showed the increase of protein content with percentages 11.20 and 14.5% in lasagna pasta of control and 7.5% algae, respectively. Phytochemicals and antioxidant activity increased with increasing algae percentage but cooking decreased all phytochemicals compared to the uncooked lasagna pasta. Cooking loss was not significantly affected with increasing algae in lasagna pasta while weight gain decreased compared to control sample. Texture analyses showed that, firmness of lasagna pasta increased with increasing the addition percentage of algae as compared to control sample. Cohesiveness, chewiness and springiness values decreased in all lasagna pasta samples compared to wheat lasagna pasta. Hunter color parameter indicated that, lightness of cooked lasagna pasta was higher than dried lasagna pasta. Also, redness (a^* values) of algae lasagna pasta had negative a^* and positive yellowness (b^*) due to the chlorophyll content. Lasagna pasta of 2.5% algae had higher color, taste, texture and overall acceptability scores compared to other higher algae content. The results recommended using wheat flour with 2.5% *S. obliquus* to improve the nutritional quality of lasagna pasta.

Keywords: *Scenedesmus obliquus*, Phytochemicals, Lasagna pasta, Quality characteristics, Texture evaluation.

INTRODUCTION

Fresh water microalgae, exceedingly distributed in rivers, lakes and polar waters exhibit a varied range of morphological, cellular, structural and biochemical composition.¹ Among these microalgal species is *Scenedesmus*, that belongs to Sphaeropleales order of Scenedesmaceae family which is frequently predominant in freshwater lakes and rivers.² Various species of this genus are used worldwide for different purposes due to their potential to adapt to harsh environmental conditions, grow rapidly and ease of cultivation and handling.^{3,4} *Scenedesmus* sp. has been used in a lot of biotechnological purposes that depend on its high nutritional content and bioactivities.^{5,6}

Enormous power for a greater diversity of microalgal species is used in human nutrition. Abundant species of microalgae contain proteins of high quality for humans.⁷ Most of common microalgal pigments such as; chlorophyll, lutein, β -carotene and phycobiliproteins are valuable to human health have anticancer, antioxidant and anti-inflammatory effects.⁸⁻¹⁰ Also in many algal species, nutritious polyunsaturated fatty acids possess essential fatty acids essential for cardiovascular, ocular and neurological health.¹¹ Regardless of this potential, few microalgal species are currently used in market for the human nutrition.

Despite, *Scenedesmus* genus is widely known as a source of food for herbivorous zooplankton and in bio fuel production due to its high oil content besides, it has exhibited the potential of being a source of high-value components with antibacterial characters. These antibacterial effects have a wide range of applications in different industries that have not been broadly investigated and fully exploited. The nutritious biochemical components of *Scenedesmus* sp., initial food acceptability, preparation, animal and human experiments indicated that, it may have a good future in health food applications. *Scenedesmus* sp. has previously been suggested for nutraceutical usage because its concentrations of essential amino acids, a good content of protein, lipid, fatty acids, eicosapentaenoic acid, essential minerals and vitamins.¹²⁻¹³

Microalgae have the ability to enhance the nutritional content of food consequently, to favorably affect humans' health because of their original chemical composition.¹⁴ *Scenedesmus* contains very high nutritional quality and several toxicological values that have not revealed any toxic impacts or abnormalities in experimental animals.¹⁵ Gross et al.¹⁶ carried out a nutritional study by incorporating *Scenedesmus* sp. into the diet of children (5 g/daily) and adults (10 g/daily). Pasta and its products are considered the main subgroup of many diets.¹⁷ It is prepared by using dough made from



any suitable material including semolina, flour of durum, farina flour, corn, rice, wheat or any mixing of these with water. In addition, pasta can be enriched, supplemented, fortified or remained conventional. Recently, several ingredients and additives have been advanced and are being utilized to improve the quality of pasta.¹⁸

It is motivating that, durum wheat (*Triticum durum*), the fundamental ingredient in "Italian style" pasta, shares only to 5% of the world's wheat production and it in general trades for a higher price than the common wheat (*Triticum aestivum*). To meet the growing pasta consumption, it is necessary to produce pasta from unconventional commodities but adequate processing and balanced formulations are required to counteract their poor technological properties.¹⁸ Therefore, the objective of this study is to investigate the possibility of using wheat flour with different percents of *S. obliquus* powder to improve the nutritional qualities of lasagna pasta.

MATERIALS AND METHODS

Samples, ingredients and chemicals

Bread wheat flour (72% extraction rate, from five stars Company, Swiss city, Egypt) and salt were purchased from local markets in Giza, Egypt. All chemicals were of analytical grade. Senile male and female mice (23-25 g) were used in the acute toxicity test. Each group of mice (8 mice each) was kept in a stainless steel cage, water and food were given ad libitum. Animal experiment was carried out according to the Medical Research Ethics Committee for institutional and national guide for the care and use of laboratory animals, National Research Centre (NRC); Cairo, Egypt.

Isolation, purification and identification of *S. obliquus*

The green alga *S. obliquus* was obtained from Algal Technology group, NRC, Dokki, Giza, Egypt. *S. obliquus* (Figure 1) was isolated from the River Nile water by using BG11 media for algal isolation and purification.¹⁹ *S. obliquus* identification has been done according to the keys of identification.¹⁹ The strain was isolated by spreading 0.1ml of water samples into petridishes containing BG11 with 1.5% agar for solidification. Single colonies of algae were then re-cultivated as nonaxenic batch cultures (50ml) at 25±2°C for 24h with continuous white fluorescent lamp intensity ≈2500Lux.



Figure 1: *S. Obliquus* microalga

BG11 nutrient composition

Nutrient composition of BG11 was used²⁰ for growing *S. obliquus* as shown in Table (1).

Table 1: BG11 nutrient composition

Nutrients	Concentration
Macronutrients	mg/l
NaNO ₃	1500.00
K ₂ HPO ₄	40.00
MgSO ₄ ·7H ₂ O	75.00
CaCl ₂ ·2H ₂ O	36.00
Citric acid	6.00
Na ₂ CO ₃	20.00
Na ₂ EDTA	1.00
Ferric ammonium citrate	6.00
Micronutrients	g/l
H ₃ BO ₃	2.86
MnCl ₂ ·4H ₂ O	1.81
ZnSO ₄ ·7H ₂ O	0.222
Na ₂ MoO ₄ ·2H ₂ O	0.39
CuSO ₄ ·5H ₂ O	0.079
Co(NO ₃) ₂ ·6H ₂ O	0.0494

1 ml/L of micronutrients solution was added to the culture

Cultivation of *S. obliquus*

The organism was grown in conical flask 5 L containing 2 L BG11 media for two weeks. After the growth phase, the algal biomass was harvested and inoculated in an open pond with capacity of 25 l and cultured with BG11 media for another 3 weeks.²⁰ The algal biomass was harvested using cooling centrifuge at 4000 rpm, and then dried at 60°C. The oil was extracted²¹, and then the algae powder was dried in oven at 70°C until complete dryness.

Analysis of ingredients and lasagna pasta

Proximate analysis

Protein, fat, crude fiber and ash contents of *S. obliquus*, wheat flour and dried lasagna pasta samples were determined as described by AOAC.²² Total carbohydrate was calculated by difference.

Phytochemicals and antioxidant activity

Half gram of algae and lasagna pasta samples were extracted with 10 ml 80% ethanol and shaken for 2 h, filtrated and the volume was made up to a known volume.

Total phenolic compounds

Total phenolic content was determined according to Singleton and Rossi²³ using Folin-Ciocalteu method. 0.25 ml of previous extract was mixed with 0.25 ml Folin-Ciocalteu reagent and 0.50 ml of 10% sodium carbonate (Na₂CO₃), then the volume was completed to 5 ml with

distilled water. After incubation in dark at room temperature for 30 min, the absorbance of the mixture was measured at 725 nm against the blank. Gallic acid was used as a standard.

Chlorophyll content

Chlorophyll a, chlorophyll b and carotenoid contents were measured using spectrophotometer according to Yang et al.²⁴ Algae and lasagna pasta (0.25 g) were homogenized with 5 ml of 85 % aqueous acetone for 5 min, then centrifuged and the supernatant was made up to known volume with 85 % acetone. The absorbance was measured at 452.5, 644 and 663 nm.

Antioxidant activity

a) DPPH free radical scavenging activity

The antioxidant activity of samples was determined according to Brand-Williams et al.²⁵ using previous extract based on the radical scavenging ability in reacting with a stable DPPH free radical. Briefly, 2.40mg of DPPH in 100 ml ethanol was prepared and 3.90 ml of this solution was added to 0.10 ml of extract. The mixture was shaken vigorously using shaker for a few second and allowed to stand at room temperature for 30 min in the dark. Then the absorbance was measured at 515nm. The DPPH radical scavenging percentage was calculated using the following equation:

$$\text{DPPH radical Scavenging activity (\%)} = \frac{A_0 - A_1}{A_0} \times 100$$

Where, A_0 = Absorbance of control (containing all reagents except the test compounds).

A_1 = Absorbance in the presence of the tested extracts after 30 min.

b) Total antioxidant capacity

One ml of *S. obliquus* and lasagna pasta with algae ethanolic extracts was mixed with 3 ml of reagent solution (0.6 M sulfuric acid, 28mM sodium phosphate and 4 mM ammonium molybdate). The tubes were capped and incubated at 95°C for 90 min. After cooling, the absorbance of each sample was measured at 695 nm. Standard series concentrations of ascorbic acid ($\mu\text{M/ml}$) were treated as the sample.²⁶

Acute oral lethal toxicity test for *S. obliquus*

Acute oral lethal toxicity test of *S. obliquus* powder was carried out according to Goodman et al.²⁷ The 24 h mortality counts (if any) among equal sized groups of mice receiving progressively increasing oral dose levels were recorded.

Amino acids determination

Amino acids composition of *S. obliquus* powder and dried lasagna pasta (control and 2.5% algae) was determined using amino acid analyzer (Biochrom 30) according to the method outlined in AOAC.²² An aliquot sample were weighed and digested with 25 ml of HCl (6 N) at 110°C for 24 h. Then, HCl was removed by evaporation; the remaining solid fraction was dissolved by 0.2 N sodium citrate buffers (pH 2.2). One ml of the solution was filtered through 0.45 μm . The standard amino acids (consist of 17 amino acids) were treated as the same as of the samples. Amino acids were expressed as g/100 g protein on dry weight basis.

Fatty acids determination

Oil was extracted from *S. obliquus* with n-hexane. The methyl esters of fatty acids were prepared according to the method of AOAC.²² Fatty acids were analyzed by gas chromatography (Agilent HP 6890) and reported in relative area percentages. The fatty acid methyl esters were identified by comparing their retention times with known fatty acid standard mixture. Peak areas were automatically computed by an integrator. The fatty acids composition was expressed as percentage of total fatty acids.

Lasagna pasta processing

Control sample of lasagna pasta was prepared using 100% wheat flour (72% extraction rate). Lasagna pasta of algae powder was prepared at three concentration levels using 2.5, 5.0 and 7.5% algae powder with wheat flour (72% extraction). 100 g wheat flour and 1.25 g salt were mixed with a definite amount of water (46-48 ml). The dough was shaped by using manual pasta maker of Titania brand (single unite steal pasta machine, Italy) and then cut into a lasagna sheets with 1.50 mm thickness, 70 mm width and 130 mm length averages. Lasagna pasta samples were dried at 45 \pm 5°C overnight, then packed in polyethylene bags and kept for further analysis at 4°C, then the most accepted lasagna pasta of algae powder in panel test (2.5%) and control sample were examined for amino and fatty acid contents.

Cooking quality of lasagna pasta

Cooking tests of lasagna pasta (weight gain and cooking loss) were performed in triplicates as following: 10 g of samples were cooked for 8-9 min in 200 ml of boiling water; the pasta was then cooled in cold water for 10 s, drained and weighed immediately. Cooking loss % (the amount of solid substance lost in the cooking water) and weight gain of different pasta samples were determined as described by Lai²⁸ then calculated according to the following equations:



$$\text{Cooking loss (\%)} = \frac{\text{weight of dried residue in cooking water}}{\text{weight of dried pasta}} \times 100$$

$$\text{Weight gain (\%)} = \frac{\text{weight of cooked pasta} - \text{weight of dried pasta}}{\text{weight of dried pasta}} \times 100$$

Cooked lasagna pasta samples were used for textural, sensorial and further analysis.

Physical properties of lasagna pasta

Color measurement

The color of dried and cooked lasagna pasta samples was measured by a hand-held Tristimulus reflectance colorimeter Minolta Chromameter (model CR-400, Konica Minolta, Japan). Results recorded in the L* a* b* c* h color system. The obtained values are presented as a means of triplicate determinations. The total color difference between dried and cooked lasagna pasta were determined according to the following equation:

$$\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2}$$

Textural profile analysis (TPA)

The texture of cooked lasagna pasta was determined by universal testing machine (Conetech, B type, Taiwan) provided with software as described by Bourne.²⁹ An aluminum 25 mm diameter cylindrical probe was used in a TPA double compression test to penetrate perpendicular to 50% depth relative to the sample height, at 1 mm/s speed test (based on pre-test). Firmness (N), gumminess (N) chewiness (N), cohesiveness and springiness were calculated from TPA graphic.

Sensory evaluation of lasagna pasta

Sensory attributes of cooked lasagna pasta were approved out by a sensory panelists from Food

Technology Research Institute according to the method of Bashir et al.³⁰ Five parameters were examined, i.e., color, aroma, taste, texture and overall acceptability using a 9-point hedonic scale.

Statistical analysis

For the analytical data, mean values and standard deviation are reported. The obtained data were subjected to one-way analysis of variance (ANOVA) at $P < 0.05$ followed by Duncan's new multiple range tests to assess differences between samples mean using Costat statistical software.

RESULTS AND DISCUSSION

Proximate analysis of wheat flour and *S. obliquus* powder

Table (2) showed the chemical composition of *S. obliquus* and wheat flour. The results indicated that, *S. obliquus* powder had high contents of protein (52.43%), fats (2.02%), crude fiber (4.53%) and ash (6.13%). While, wheat flour (72% extraction rate) had a high content of total carbohydrate (86.74%). The current results are in agreement with the results of Batista et al.³¹ and Becker³² who found that, *S. obliquus* had 50 to 56% protein and 33 to 64% total carbohydrate, respectively. For wheat flour, the present results are close to the results reported by Seleem and Omran.³³

Table 2: Proximate analysis of *S. obliquus* powder and wheat flour (% on dry weight basis)

Ingredients	<i>S. obliquus</i> powder	Wheat flour (72% extraction rate)
Protein	52.43 ± 0.98 ^a	11.30 ± 0.41 ^b
Fats	2.02 ± 0.12 ^a	0.91 ± 0.09 ^b
Crude fiber	4.53 ± 0.50 ^a	0.42 ± 0.03 ^b
Ash	6.13 ± 0.57 ^a	0.63 ± 0.07 ^b
Total carbohydrate*	34.89 ± 1.52 ^b	86.74 ± 0.55 ^a

Each value is expressed as mean of three replicates ± SD, number in the same column followed by the same letter are not significantly different at 0.05 level. * Total carbohydrate was calculated by differences.

Phytochemical contents, antioxidant activity and lethal toxicity of *S. obliquus*

Table (3) illustrates the pigment contents of *S. obliquus*. The results indicated that, *S. obliquus* powder characterized with its high phenolic compounds (2333.08 mg/100g). Also, *S. obliquus* powder had 699.90, 1437.50 and 568.80 mg/100g for chlorophyll a, chlorophyll b and carotenoids, respectively. While, DPPH free radical scavenging activity was 45.27% and total antioxidant capacity was 298.85 µg/ml. These results are in the same

trend with the results of Kent et al.³⁴ who found that, *Scenedesmus* sp. had 1900 mg/100g chlorophyll, 423.0 mg/100g carotenoids. Moreover, the acute lethal toxicity test revealed that, *S. obliquus* was very safe up to the highest tested dose (10g/kg mice body weight). This dose corresponds to 78 g/70 kg man body weight for human while the dose of mice was extrapolated to corresponding estimates in human adopting interspecies dosage conversion scheme.³⁵ This reflects the highest safety of *S. obliquus*.



Amino acids content and amino acid scores of algae and lasagna pasta

Amino acid contents was evaluated in *S. obliquus* powder, lasagna pasta of 2.5% algae and compared with lasagna of control sample as shown in Table (4).

Table 3: Phytochemical contents, antioxidant activity and lethal toxicity of *S. obliquus* powder

Constuites	Content (mg/100g)
Total phenolic	2333.08±16.14
Chlorophyll a	699.90 ± 5.50
Chlorophyll b	1437.50 ± 6.10
Carotenoids	568.80 ± 5.50
DPPH free radical scavenging activity (%)	45.27 ± 1.80
Total antioxidant capacity (µg/ml)*	298.85 ± 1.01
Lethal toxicity	10 g/kg mice body weight

*Total antioxidant capacity (µg/ml) as ascorbic acid. Each value is expressed as mean of three replicates ±SD.

The obtained results indicated that, algae powder contains higher amounts of total essential amino acids especially, leucine which was the most abundant essential amino acid in *S. obliquus* powder followed by lysine and threonine. Also, control sample of lasagna pasta (100% wheat) contained high amounts of total nonessential amino acids especially glutamic acid that was the most abundant nonessential amino acid in wheat. Lasagna pasta of 2.5% *S. obliquus* powder contained higher amount of total essential amino acids while total non essential amino acids decreased as compared to control sample of lasagna pasta. The results are close to those findings by Kent et al.³⁴ for microalgae and commercial microalgae products. Also, Spolaore et al.¹¹ stated that, protein content (both in amino acids content and essential amino acid supply for body) of most algae is higher than that of other protein foods.

Table 4: Amino acids content of *S. obliquus* powder, lasagna pasta of control and *S. obliquus*

Amino acids g/ 100g protein	<i>S. obliquus</i> powder	Lasagna pasta (control)	Lasagna pasta with 2.5% <i>S. obliquus</i>
Essential amino acids			
Isoleucine	3.53±0.07 ^a	2.68±0.05 ^b	2.79±0.08 ^b
Leucine	7.63±0.14 ^a	5.44±0.11 ^b	5.01±0.15 ^c
Lysine	5.38±0.10 ^a	2.06±0.04 ^c	2.46±0.07 ^b
Methionine	2.17±0.04 ^a	2.05±0.03 ^b	2.05±0.06 ^b
Phenylalanine	5.19±0.09 ^a	4.91±0.11 ^b	4.92±0.14 ^b
Threonine	5.36±0.10 ^a	2.59±0.05 ^c	3.28±0.10 ^b
Valine	5.36±0.10 ^a	4.19±0.08 ^b	4.35±0.13 ^b
Histidine	1.72±0.03 ^b	2.32±0.05 ^a	2.29±0.06 ^a
Nonessential amino acids			
Tyrosine	3.82±0.07 ^a	3.57±0.07 ^b	3.27±0.09 ^c
Arginine	6.49±0.12 ^a	4.10±0.05 ^c	4.34±0.11 ^b
Alanine	9.58±0.18 ^a	3.03±0.06 ^c	3.69±0.11 ^b
Aspartic acid	8.39±0.16 ^a	4.46±0.09 ^c	4.75±0.12 ^b
Cysteine	1.53±0.03 ^c	3.12±0.06 ^a	2.62±0.07 ^b
Glutamic acid	10.53±0.20 ^c	33.72±0.66 ^a	31.77±0.83 ^b
Glycine	5.38±0.11 ^a	3.57±0.16 ^b	3.68±0.10 ^b
Proline	4.24±0.08 ^b	9.81±0.19 ^a	9.82±0.26 ^a
Serine	4.39±0.07 ^b	3.92±0.08 ^c	4.75±0.12 ^a

Each value is expressed as mean of three replicates ±SD, number in the same row followed by the same letter are not significantly different at 0.05 level.

Amino acid scores of previous samples were calculated and presented in Table (5). The obtained results showed that, *S. obliquus* had a higher percent of amino acid scores compared to control lasagna pasta and lasagna pasta of 2.5% *S. obliquus* powder. Result agreed with Kent et al.³⁴ who reported that, amino acid scores were higher in microalgae principally due to higher proportions of the essential amino acids.

Fatty acids composition

Table (6) presents percentages of fatty acids of algae, control and 2.5% lasagna pasta with algae. The results indicated that, algae had a high content of fatty acids especially C8:0, C10:0, C12:0, C13:0, C14:0, C16:1, C17:0, C17:1, C18:0, C18:2, C18: 3n3, C18:3n6, C20:0 and C20:1. From the data in Table (6) it could be stated that, control pasta is rich in C16:0, C18:0, C18:1 and C18:2. The



dominant fatty acids were C16:0, C18:1 for algae; C16:0, C18:1, C18:2 for control and 2.5% lasagna pasta. The results are close to those findings by Custodio et al.³⁶ and Kent et al.³⁴

Table 5: Amino acid scores of *S. obliquus*, lasagna pasta of control and lasagna pasta mixed with *S. obliquus*

Amino acids	FAO/WHO Reference value*	Amino acid scores (%)		
		<i>S. obliquus</i> powder	Lasagna pasta(control)	Lasagna pasta of 2.5% <i>S. obliquus</i>
Isoleucine	6.60	126.07	95.71	99.64
Leucine	2.80	115.61	82.42	75.91
Lysine	5.80	92.76	35.52	42.41
Methionine+Cysteine	2.50	148.00	206.80	186.80
Phenylalanine +Tyrosine	6.30	143.02	134.60	130.00
Threonine	3.40	157.65	76.18	96.47
Valine	3.50	153.14	119.71	124.29
Histidine	1.90	90.53	122.11	120.53
Total amino acids	32.80	127.10	100.40	100.73

*FAO/WHO 1991. Protein quality evaluation; Food and Agriculture Organization of the United Nations. Daily requirements for human adults.1991, Rome, Italy.P. 66.

Table 6: Fatty acids content of *S. obliquus* powder, lasagna pasta of control and algae

Fatty Acid(% of total fatty acids)	S. obliquus powder	lasagna pasta of	
		100% wheat	2.5% S. obliquus
Saturated fatty acids			
C8:0	6.93	0.71	2.56
C10:0	1.21	0	0
C12:0	6.3	0	1.03
C13:0	5.33	0.09	0.27
C14:0	5.22	0.23	0.56
C15:0	0.2	0.11	0.15
C16:0	16.26	18.62	16.53
C17:0	1.09	0.43	0.66
C18:0	2.97	5.09	2.75
C20:0	1.57	0.68	0.45
C22:0	0.27	0.69	2.56
Subtotal	47.35	26.65	27.52
Mono unsaturated fatty acids			
C14:1	0.16	0	0
C15:1	0	0.19	0
C16:1	1.38	0.31	0.53
C17:1	6.22	0.06	0.61
C18:1	7.87	19.62	16.33
C20:1	1.29	0.75	0.5
C22:1	-	-	-
Subtotal	16.92	20.93	17.97
Poly unsaturated fatty acids			
C18: 2	16.83	49.46	48.62
C18: 3n3	14.13	2.33	3.76
C18:3n6	4.76	0.23	1.85
Subtotal	35.73	52.42	54.23

Chemical composition of lasagna pasta

Chemical composition of lasagna pasta(control) and lasagna pasta that mixed with 2.5, 5 and 7.5% *S. obliquus* powder are presented in Table (7). The results indicated that, increasing algae percentage in lasagna pasta significantly increased protein, ash and crude fiber contents compared to control lasagna pasta. This result could be due to, the high content of protein and ash in *S. obliquus*. Regarding to fat contents, Table (7) showed that lasagna pasta not affected with different addition levels of algae. While, total carbohydrate was significantly decreased with increasing level of algae.

Fard et al.³⁷ declared that, the addition of microalga to cookies significantly increased the protein content of fortified samples as compared with control. Spirulina may be used in bakery products for protein enrichment without any significant changes in the texture.³⁸ Also Fard et al.³⁷ stated that, the fortified cookies with Spirulina at the concentrations of 0, 0.5, 1 and 1.5% (w/w) to the cookies formulation led to significantly higher protein content. Also Abo El-Naga et al.³⁹ stated that, supplementation of pasta with high protein source like surimi and mushroom resulted in a better quality and nutritious.

Table 7: Proximate chemical content of lasagna pasta mixed with *S. obliquus* (%dry weight basis)

Lasagna pasta	Protein	Fats	Ash	Crude fiber	Total carbohydrate
Control	11.20±0.24 ^d	0.86±0.04 ^a	1.05±0.03 ^c	0.40±0.02 ^d	86.75±0.39 ^a
2.5% <i>S. obliquus</i>	12.22±0.32 ^c	0.88±0.02 ^a	1.11±0.05 ^c	0.50±0.02 ^c	85.79±0.34 ^b
5.0% <i>S. obliquus</i>	13.34±0.11 ^b	0.90±0.05 ^a	1.28±0.04 ^b	0.56±0.04 ^b	84.48±0.19 ^c
7.5% <i>S. obliquus</i>	14.50±0.16 ^a	0.93±0.03 ^a	1.46±0.06 ^a	0.63±0.02 ^a	83.11±0.23 ^d

*Total carbohydrate was calculated by differences. Each value is expressed as mean of three replicates ±SD, number in the same column followed by the same letter are not significantly different at 0.05 level.

Phytochemical contents and antioxidant activity of lasagna pasta before and after cooking

Table (8) showed phytochemical contents of lasagna pasta before and after cooking. The results indicated that, phytochemicals and antioxidant activity increased with increasing the mixing level of algae. On the other hand, cooking of lasagna pasta decreased phytochemicals compared to dried lasagna pasta and this could be due to the leach in cooking water. Suman et al.⁴⁰ mentioned that, dried lasagna samples showed a drastic decrease in

the associated chlorophylls in addition to, a simultaneous increase in pheophytine and pheophorbide content, mainly due of the thermal treatment. Turkmen et al.⁴¹ found that, cooking resulted in a loss of chlorophyll a and b to various extent depending on the type of method and the vegetable type. Besides pigment diffusion into the cooking water, pasta color losses may also be attributed to the microalgae pigment oxidation during thermal treatment.³¹

Table 8: Phytochemicals content and antioxidant activity of lasagna pasta mixed with *S. obliquus* before and after cooking

	Lasagna pasta	Lasagna pasta mixed with algae at level		
		2.5 %	5.0 %	7.5 %
Dried lasagna pasta				
Total phenolic (mg/100g)	89.41±8.61 ^d	137.01±0.60 ^c	151.97±0.24 ^b	180.02±1.73 ^a
Chlorophyll a (mg/100g)	0.00±0.00 ^d	46.90±0.07 ^c	83.8±0.91 ^b	112.5±0.55 ^a
Chlorophyll b (mg/100g)	0.00±0.00 ^d	143.0±0.9 ^b	176.10±0.52 ^a	204.30±3.20 ^a
Carotenoids (mg/100g)	0.72±0.02 ^d	67.40±0.05 ^c	82.80±0.27 ^b	196.40±5.50 ^a
DPPH radical scavenging activity (%)	9.20±0.52 ^d	17.74±4.32 ^c	26.03±1.98 ^b	35.84±4.41 ^a
Total antioxidant capacity (µg/ml) *	85.06±2.68 ^d	103.63±6.4 ^c	128.5±3.14 ^b	168.69±16.8 ^a
Cooked lasagna pasta				
Total phenolic (mg/100g)	21.71±3.88 ^d	103.94±3.55 ^c	120.16±1.18 ^b	141.48±1.37 ^a
Chlorophyll a (mg/100g)	0.00±0.00 ^c	22.0±0.60 ^b	28.70±0.60 ^b	44.50±0.60 ^a
Chlorophyll b (mg/100g)	0.00±0.00 ^d	71.4±0.49 ^c	143.0±0.54 ^b	188.40±6.0 ^a
Carotenoids (mg/100g)	0.70±0.01 ^c	18.20±0.6 ^c	38.0±0.55 ^b	53.70±0.59 ^a
DPPH radical scavenging activity (%)	5.40±0.22 ^c	13.03±2.19 ^b	20.29±0.61 ^a	24.99±2.52 ^a
Total antioxidant capacity (µg/ml)*	76.61±1.39 ^b	91.66±6.65 ^b	115.95±4.28 ^a	135.0±16.07 ^a

Each value is expressed as mean of three replicates ±SD, number in the same row followed by the same letter are not significantly different at 0.05 level.*as ascorbic acid



Cooking quality

Table (9) shows the effect of different mixing level of algae on cooking quality parameters of lasagna pasta (cooking losses and weight gain). Results indicated that, cooking losses of lasagna decreased slightly in mixed lasagna pasta with algae at different mixing levels compared to control sample. Also, there was no significant difference between lasagna pasta of different mixing levels with algae. The algae incorporation may weaken the structure of pasta due to their proteins are not able to develop a gluten network and steric hindrance interferes in network formation. Due to the protein network limits the water diffusion into the central zone of pasta during cooking⁴², weaker gluten-protein network facilitates water diffusion into it, reducing cooking time. In addition, since the gluten-protein network is responsible for retaining pasta a physical integrity during cooking, a weaker structure leaches more solids from pasta samples into cooking water, increasing cooking residues.^{43, 44} On the other hand, cooking loss values in all samples were less than 8% which is the limit value considered desirable for wheat pasta as mentioned by Dick & Youngs⁴⁵ who stated that, cooking losses are a serious indicator for the pasta cooking performance by both consumers and industry.

Higher weight gain values of the samples enriched with microalgae indicated that, they absorbed higher amounts of water during cooking. The addition of *S. obliquus* (2.5, 5.0 and 7.5%) in lasagna pasta induced a significant decrease of weight gain (66.33, 69.10 and 76.53%) compared with control lasagna pasta (107.12%). Pasta prepared with *S. obliquus* presents weight gain lower than the control lasagna pasta. This result may be attributed to the ability of *S. obliquus* to absorb water and retain it in the protein-starch net. Accordingly, weight gain was also affected by the utilization of *S. obliquus* biomass, which decreased from 107.12% in the control pasta to 76.53% in 7.50% algae powder. Furthermore, these results are in accordance with those obtained by Zardetto & Rosa⁴⁶ who reported that, the pasta prepared with 20% of microalgae possessed higher water absorption than other samples reported. Although, the lack of a continuous protein network causes high hydration of the starch material and increases the weight of pasta. When gluten was diluted by the substitution of wheat flour to spirulina biomass, the protein network became weaker, facilitating the leak of amylose into cooking water.⁴⁷

Table 9: Cooking quality of wheat and lasagna pasta with *S. obliquus* (%)

Lasagna pasta	Cooking loss	Weight gain
Control	5.62±0.31 ^a	107.12±2.98 ^a
2.5% <i>S. obliquus</i>	5.17±0.35 ^b	66.33±4.70 ^b
5.0% <i>S. obliquus</i>	5.27±0.08 ^{ab}	69.10±2.03 ^b
7.5% <i>S. obliquus</i>	5.48±0.25 ^{ab}	76.53±2.50 ^b

Each value is expressed as mean of three replicates ±SD, number in the same column followed by the same letter are not significantly different at 0.05 level.

Physical properties

Texture Profile Analysis (TPA)

Color

Table (10) displays color parameters (L, a, b, c, h and ΔE) of dried and cooked lasagna pasta prepared from algae powder. It could be noticed from these results that, substitution of wheat flour with algae in the formulation of lasagna pasta affected the lasagna pasta color. Data in Table (10) also indicated that, substituting of wheat flour with *S. obliquus* significantly decreased the lightness (L) values of dried and cooked lasagna pasta with increasing level of substitution where, control lasagna pasta recorded the highest value. However, L values increased in cooked lasagna pasta compared with dried lasagna pasta and became lighter (L value increases). The a values of lasagna pasta are agreed with Zouari et al.¹⁴ results who found that, firmness of pasta containing *A. platensis* had negative a and positive b due to the chlorophyll content of these algae. During the cooking of lasagna pasta, there is a tendency for the color to

become less green (a becomes less negative) and more yellow (b becomes more positive).⁴⁰

Regarding, c (color saturation) and h values of lasagna pasta were found to be increased with increasing level of substitution. Besides, *S. obliquus* lasagna pasta had the highest hue angle values as compared to control pasta. These results are in agreement with Zouari et al.¹⁴ who found that, after cooking, all samples of pasta enriched with *A. platensis* microalgae were slightly brighter and more yellow. Color losses resulting after cooking of pasta are expressed in terms of total color difference (ΔE) between dried and cooked lasagna pasta as specified in Table (10). The substitution with algae with 2.5, 5.0 and 7.5% had a higher color loss and this may be due to the thermal process which has affected the color. Also, pasta color losses arise from pigment leaching into the cooking water and may also be related to *S. Obliquus* pigment oxidation during thermal treatment. Microalgae are recognized as an excellent source of pigments including; chlorophylls, carotenoids and phycobiliproteins. This fact explained the reduction in color parameter values.¹⁴



Table 10: Color of dried and cooked lasagna pasta mixed with *S. obliquus*

Lasagna pasta samples Parameters	Lasagna pasta	Lasagna pasta mixed with <i>S. obliquus</i> at levels		
		2.5%	5.0%	7.5%
L				
Dried	73.97±2.12 ^a	38.29±1.70 ^b	33.53±0.16 ^c	29.38±0.33 ^d
Cooked	77.41±0.97 ^a	41.68±0.97 ^b	39.06±0.19 ^b	31.94±1.69 ^c
a				
Dried	0.10±0.03 ^a	-3.83±0.09 ^d	-2.32±0.02 ^c	-1.26±0.04 ^b
Cooked	-2.23±0.11 ^b	-4.11±0.19 ^d	-3.61±0.06 ^c	-1.57±0.08 ^a
b				
Dried	14.73±0.28 ^a	10.59±0.43 ^b	7.56±0.36 ^c	5.63±0.17 ^d
Cooked	19.46±0.64 ^a	17.43±0.38 ^b	16.74±0.31 ^{bc}	16.61±0.11 ^c
c				
Dried	14.80±0.24 ^a	11.26±0.43 ^b	7.91±0.35 ^c	5.77±0.16 ^d
Cooked	19.58±0.63 ^a	17.91±0.33 ^b	17.13±0.29 ^c	16.69±0.10 ^c
h				
Dried	89.62±0.09 ^d	109.92±0.28 ^a	107.08±0.63 ^b	102.63±0.65 ^c
Cooked	96.54±0.42 ^c	103.28±0.87 ^a	102.18±0.42 ^b	95.40±0.26 ^d
ΔE	-	7.82±1.37 ^b	11.09±0.42 ^a	11.36±0.43 ^a
Color	Yellow	Greenish yellow	Greenish yellow	Greenish yellow

*L (lightness with L = 100 for lightness, and L = zero for darkness), a [(chromaticity on a green (-) to red (+))], b [(chromaticity on a blue (-) to yellow (+))], c (color saturation), h [(hue angle where 0° = red to purple, 90° = yellow, 180° = bluish to green and 270° = blue scale. ΔE is the total color difference between dried and cooked lasagna pasta. Each value is expressed as mean of three replicates ±SD, number in the same row followed by the same letter are not significantly different at 0.05 level.

Texture profile analysis of cooked lasagna pasta

Table (11) demonstrates textural profile (firmness, cohesiveness, gumminess, chewiness and springiness) of cooked lasagna pasta prepared from *S. obliquus* powder. Depending on the present results, substitution of wheat flour with *S. obliquus* powder in formulation of lasagna pasta affected the lasagna pasta textural properties. Values of lasagna pasta firmness and gumminess increased with increasing the substitutions percent of *S. obliquus* powder as compared to wheat lasagna pasta.

The current results are in agreement with Zouari et al.¹⁴ who found that, firmness of pasta containing *Arthrospira platensis* appeared to be higher than that of the wheat control pasta. Besides, there was a decrease in cohesiveness, chewiness and springiness values in all lasagna pasta samples compared to wheat lasagna pasta. Moreover De Marco et al.⁴⁷ found that, replacing the wheat flour with spirulina led to an increase of firmness, cohesiveness and chewiness.

Table 11: Texture profile analysis of cooked lasagna pasta mixed with *S. obliquus*

Lasagna pasta samples	Firmness(N)	Cohesiveness	Gumminess (N)	Chewiness(N)	Springiness
Control	1.88±0.02 ^d	0.61±0.01 ^a	1.15±0.10 ^b	0.98±0.08 ^a	0.85±0.03 ^a
2.5% <i>S. obliquus</i>	2.23±0.03 ^c	0.57±0.03 ^b	1.27±0.09 ^a	0.96±0.07 ^a	0.75±0.02 ^b
5.0% <i>S. obliquus</i>	2.37±0.03 ^b	0.56±0.02 ^b	1.33±0.03 ^a	0.94±0.08 ^a	0.71±0.03 ^c
7.5% <i>S. obliquus</i>	2.50±0.05 ^a	0.54±0.04 ^b	1.35±0.03 ^a	0.82±0.03 ^b	0.61±0.08 ^d

N= Newton. Each value is expressed as mean of three replicates ±SD, number in the same column followed by the same letter are not significantly different at 0.05 level.



Sensory characteristics

The values in Table (12) indicate the percentage of acceptability in each attribute; color, aroma, texture, taste and overall acceptability. Lasagna pasta was prepared by mixing wheat flour (72% extraction rate) with 2.5, 5.0 and 7.5% algae *S. obliquus* powder. Results in Table (12) showed that, substituting of wheat flour with 2.5% microalgae had higher color, taste, texture and overall acceptability scores compared to other substitution percents (5.0 and 7.5%).

On the other hand, substitution of wheat flour with *S. obliquus* microalgae at 5.0 and 7.50% significantly

decreased sensorial parameters. In addition, arome score significantly decreased in all samples compared to control. It can be noticed that, *S. obliquus* could be incorporated up to 2.5 g/100 g of wheat flour in the formulation of lasagna pasta without any effect on acceptability. These results are close to those of Zouari et al.¹⁴ who mentioned that, *A. platensis* may incorporated up to 2 g/100 g of semolina in pasta formulation with enhancing its sensory quality due to the microalgal effects on brightness of the cupcakes. Also, Danesi et al.³⁸ reported that, bakery products may be enriched with *Spirulina* without any reverse effect on sensory acceptance of the product.

Table 12: Effect of mixing lasagna pasta with *S. Obliquus* on sensory evaluation

Lasagna pasta samples	Color	Aroma	Texture	Taste	Overall acceptability
Control	8.80±0.42 ^a	8.50±0.53 ^a	8.70±0.48 ^a	8.60±0.52 ^a	8.55±0.50 ^a
2.5% <i>S. obliquus</i>	8.00±1.03 ^{ab}	7.60±0.84 ^b	8.20±0.59 ^{ab}	7.71±0.42 ^a	7.65±0.78 ^a
5.0% <i>S. obliquus</i>	7.00±1.33 ^{bc}	6.80±1.11 ^{bc}	7.80±0.79 ^b	6.15±1.18 ^b	6.60±1.10 ^b
7.5% <i>S. obliquus</i>	6.05±1.69 ^c	6.10±1.31 ^c	7.55±1.17 ^b	5.25±1.51 ^b	5.60±1.43 ^c

Each value is expressed as mean of ten replicates ±SD, number in the same column followed by the same letter are not significantly different at 0.05 level.

CONCLUSION

The results of this study clearly demonstrated the usefulness of mixing *S. obliquus* powder with lasagna pasta as a valuable food additive to enhance the nutritional characteristics of lasagna pasta. Where, algae are a suitable source of protein, unsaturated fatty acids and pigments with a good source of essential amino acids and antioxidants effects. The most acceptable mixing level with the highest acceptability was a 2.5% *S. obliquus* powder.

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