

Quality Characteristics of Croissant Stuffed with Imitation Processed Cheese Containing Microalgae *Chlorella vulgaris* Biomass

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Abstract: Microalgae are able to enhance the nutritional content of conventional foods and hence to positively affect human health, due to their original chemical composition. The aim of the present study was to prepare fresh croissant filled with imitation cheese enriched with different amounts of *Chlorella vulgaris* biomass and to assay the quality parameters of the resulted croissant. Chemical composition of algal biomass, imitation cheese and croissant, as well as texture of cheese, staling rate, yeasts and moulds counts and sensory evaluation of resulted croissants were determined. The incorporation of microalgae resulted in an increase of quality parameters of croissant treatments when compared to the control sample. The chemical composition and staling rate of croissant remained relatively stable with the addition of cheese enhanced with microalgae. Results from sensory analysis of croissants filled with imitation cheese enhanced with different quantity of *Chlorella vulgaris* biomass showed that the using of these microalgae in imitation cheese improved the sensory quality characteristics of croissants. Use of 2 and 3 % of the microalgae significantly ($P \leq 0.05$) impact on better croissants taste, texture and eating quality. The results obtained are in accordance with the aim of this work to develop innovative croissant that are attractive to consumers with similar texture and flavor profiles to those of the traditional common croissant.

Key words: Green algae • *Chlorella vulgaris* biomass • Imitation cheese • Croissant • Quality properties

INTRODUCTION

Microalgae have a great potential for various applications including the production of compounds for food, feed and aquaculture; also of higher value products for pharmaceutical and cosmetic industries [1]. Microalgae are an enormous biological resource, representing one of the most promising sources for new food products [2]. These microscopic organisms can be grown under certain controlled environmental conditions in large amounts. The possibility of not only harvesting microalgae but also growing them at different conditions enables its use as natural reactors at a large scale [3]. Therefore, microalgae can be used to enhance the nutritional value of food products, due to their well balanced chemical composition as well as a source of highly valuable molecules, such as polyunsaturated fatty acids, pigments, sterols, vitamins, hydrocolloids and other biologically active compounds. Microalgae

production has greatly increased in recent years, with a biomass production raised from about 3500 tons per year in 2004 to about 10,000 tons per year nowadays [1]. *Chlorella vulgaris* is microscopic single celled freshwater green algae, which has health benefits, such as assisting disorders such as gastric ulcers, wounds, constipation, anemia, hypertension, diabetes, infant malnutrition and neurosis [4]. It is also considered to be a biological response modifier, as demonstrated by its protective activities against viral and bacterial infections in normal and immunosuppressed mice as well as against tumors. It is reported to be a rich source of antioxidants such as lutein, α - and β -carotene, ascorbic acid and tocopherol and supplies large quantities of minerals and dietary fibers [5]. *Chlorella vulgaris* biomass also represent a valuable source of nearly all essential vitamins (e.g., A, B1, B2, B6, B12, C, E, nicotinate, biotin, folic acid and pantothenic acid [2]. In addition to the health aspects, *Chlorella vulgaris*

is also important as a source of natural pigments, namely carotenoids and can be used as a natural coloring agent [4].

Cheese is an extremely versatile food product that has a wide range of textures, flavor and end uses. As processed cheeses (imitation cheeses) are used mainly as an ingredient in prepared foods including appetizers, soups, sauces, fillings in pastry and pies, burgers, pizzas and toasted sandwiches where it is used to add taste, texture and nutrition [6, 7]. However, because of the high costs associated with natural cheese production and storage, the industry seeks alternatives. Attempts to reduce cheese costs have led to the development of imitation cheeses based on casein and its derivatives and use the vegetable fat to replace the more costly milk fat. In addition, the manufacture of an imitation cheese allows manufacturers greater scope in manipulating constituents toward nutritional, textural and economic ends [8]. Imitation cheese is generally manufactured by blending casein or its derivatives with various edible oils/fats, proteins, other ingredients and water into a smooth homogeneous blend with the aid of heat, mechanical shear and emulsifying salts and is used as a cost effective substitute for natural [9, 10]. Rennet casein is the most often used source of protein in production of processed cheese imitations but recently acid casein was used too [11]. Bread and other bakery products like croissant have an important role in human nutrition. Therefore, there have been many investigations to enhance the nutritive value of these products to produce tasteful croissant-type products filled with various kinds of cream or cheese which are favored especially by children population; these are more and more frequently subjected to different technological treatments prolonging their durability (ingredients, packaging) as mentioned by Hozova *et al.* [12]. Croissants could be described as delicious bakery products with characteristic laminated aerated-flaky structure formed by enveloping a sheet of butter or margarine in yeast dough, folding it to increase the numbers of layers to obtain a layered structure with thin fat/dough layers. Croissants are usually consumed for breakfast or lunch [13].

The objectives of this study were to use *Chlorella vulgaris* biomass as a new functional ingredient at different levels (1, 2 and 3%) in the preparation of imitation cheeses and to use these cheeses as filling agents in croissant production, to be healthier and more attractive. The effect of these filling agents on quality properties of resultant croissant was also studied.

MATERIALS AND METHODS

Materials: *Chlorella vulgaris* biomass in the form of freeze-dried was obtained from Algal Biotechnology Unit, National Research Center, Giza, Egypt. Calf rennet powder (Ha-La) was obtained from CHR- Hansen's Lab. Denmark. Sodium chloride and citric acid were purchased from Sigma (Germany). Fresh raw buffalo's milk was obtained from the herd of the Fac. Agric., Ain Shams Univ., Cairo, Egypt. The following ingredients were purchased from local market: wheat flour (72% extraction), corn oil, margarine, eggs, milk and baking powder.

Methods of Processing

Preparation of Rennet Casein: The rennet casein was prepared according to the method described by Morr [14]. Skim milk used for preparation rennet casein and was pasteurized at 72°C for 15-20 seconds, then cooled to about 35°C; 0.01 % calcium chloride, 1.0% rennet were added and mixed well. After 1 hr at 35°C the milk was coagulated into a gel-like structure, the curd was cut manually into pieces of approximately 1 cm. The coagulum was stirred while being heated to about 60°C, the cooking time was approximately 30 minutes. The whey was drained off and the remaining casein while in the vat was washed twice or three times with water to remove whey proteins, lactose and salt at a temperature of 60°C. After the water was drained off, the casein was further dewatered in separator and then dried at 50°-55°C until the moisture content was 12% and finally ground to a powder. The proximate composition of prepared rennet casein (pH 6.62) is shown in Table 1.

Manufacture of Imitation Cheese Slices: Imitation cheese was manufactured as described by Savello *et al.* [15] and the modifications of Ye and Hewitt [6]. All experimental samples were formulated to yield imitation process cheese slices with 40% moisture and 50% fat-in-dry-matter. A control imitation cheese was manufactured using the following ingredients: 25.0% rennet casein, 30.0% corn oil, 2.5% emulsifying salts, 1.75% sodium chloride, 0.7% citric acid and 40.0 % water. The dry ingredients

Table 1: Proximate composition (% on dry basis) of rennet casein

Component	%
Moisture	12.40
Protein	80.00
Fat	1.30
Ash	6.25

and oil (except citric acid) were blended with water and hydrated at room temperature for 1h. Then placed into the processing batch type kettle of 10 kg capacity, a pilot machine locally made in Egypt at the National Research Center. All blends were cooked with controlled agitation for 3 min at 90°C using direct injection steam at pressure of 1.5 bars, citric acid was added and the blend was cooked for further 5 min. The sample was cut into a slice with uniform thickness of approximately 2.4 mm and sealed in a plastic bag. The slices were immediately cooled and stored at 4°C for further analysis. Using the similar process, three imitation cheese products containing 1, 2 and 3% of *Chlorella vulgaris* biomass were prepared by replacing equivalent quantities of rennet casein in the standard formulation with *Chlorella vulgaris* biomass which were added at the end of cooking time. All samples were adjusted at pH 5.8±0.1 by adding citric acid.

Processing of Croissant: Wheat flour (125 g) was mixed well within 50 g margarine, then 1 tsp baking powder was added. One egg was cracked into a cup and whisked; 50 ml milk was added to the cup and whisked together, then the mixture was poured into the bowl and mixed to form dough. The dough was divided in two pieces, each piece was rolled into a round shape, then divided into 4 pieces and each part was rolled up. The rolled piece were put on a baking tray, brushed with the egg and milk mixture and baked in the oven at 205°C for 10-15 min [16]. The imitation cheese enhanced with *Chlorella vulgaris* biomass at levels of 1%, 2% and 3% were added to the baked croissant as filling materials. Then, the croissant samples were wrapped with stretch film and stored at room temperature for six days.

Analytical Methods:

Analysis of Algae: Microalgae biomass nutrient profile was determined by AOAC standard methods [17] in terms of moisture, ash, protein, fat and fiber contents; total carbohydrate content was calculated by differences. Fatty acids profile were determined using gas chromatography (GC Hewlett Packard 6890) according to the methods described by AOAC [18]. For amino acid pattern, the analysis was carried out as described by AOAC [17] using an Eppendorf Biotronic LC 3000 Amino acid Analyzer (Eppendorf- Biotronic, Hamburg, Germany).

Analysis of Imitation Cheeses: Imitation cheese slices were tested for fat content using Gerber method, total protein using micro-Kjeldahl method and pH values

using pH meter model Cole-Armer Instrument Co., USA as described in Ling [19]. The salt content was determined as described by Bradley *et al.* [20]. The moisture and ash contents were determined according to the method in AOAC [17].

Analysis of Croissant: Moisture, protein, fat and ash contents of different croissant samples were determined according to AACC [21]. The staling rate of prepared croissant was determined after baking within one hour and after 3 and 6 days of storage at room temperature (18±2°C) by alkaline water retention capacity (AWRC %) using the method of AACC [21]. Yeast and mould counts were determined by the plate count method according to AACC [21] for croissant samples during storage period.

Sensory Evaluation: Sensory evaluation of fresh and stored croissant at 18±2°C was determined according to Bennion and Bamford [22]. The evaluation was done by ten of the staff members at Department of Food Science, Fac. Agric., Ain Shams Univ., Cairo, Egypt. Croissant samples were scored as follows, crust color (20), taste/odor (25 points), texture (15 points), crust appearance (15 points) as well as volume and eating quality (25 points).

Statistical Analysis: Statistical analysis was carried out using ANOVA procedures followed by Duncan's Multiple Range Test with $P \leq 0.05$ being considered statistically significant to compare between means according to Snedecor and Cochran [23]. All procedures were in triplicate using Statistical Analysis System program [24].

RESULTS AND DISCUSSION

Proximate Composition of *Chlorella Vulgaris* Biomass:

Table 2 show the proximate composition of *C. vulgaris* biomass which used in this study (pH 6.50). The results revealed that the protein content was accounted about half the solids of the biomass (51.45%). The other components Carbohydrates, fat, fiber and ash represented 11.86, 12.18, 9.18 and 9.50, respectively. The contents of protein and ash in algae are in agreement with those obtained by Janczyk *et al.* [25] who mentioned that the protein and ash contents were 52.8 and 9.13 %, respectively, while they found that the contents of fat and carbohydrates were less than our results (8.1 and 5.6 %, respectively), despite the fiber content was so high (20.8%). These differences could be due to

Table 2: Proximate composition (% dry basis) of *Chlorella vulgaris* biomass

Component	%
Moisture	5.83
Crude protein	51.45
Carbohydrates*	11.86
Fat	12.18
Crude fiber	9.18
Ash	9.50

*: calculated by differences

Table 3: Fatty acid composition of oil extracted from *Chlorella vulgaris* biomass (% of TFA)

Fatty acid	Name	(%)
C _{14:0}	Myristic	1.85
C _{16:0}	Palmitic	52.56
C _{17:1}	Heptadecanoic	15.88
C _{18:0}	Stearic	8.83
C _{18:1}	Oleic	5.42
C _{18:2}	Linoleic	6.56
C _{18:3}	Linolenic	8.87
TSFA*	-	63.24
TUSFA**	-	36.73

TSFA: total saturated fatty acids.

TUSFA: total unsaturated fatty acids.

the differences in cultivation methods of one strain of algae as mentioned by Janczyk *et al.* [26]. *Chlorella vulgaris* is sometimes called as the “whole food” as it contains loads of nutrients which are essential to human health. *Chlorella vulgaris* contained the highest amount of minerals compared to other green algae. This is in correlation with the higher amount of ash as evident from Table 2. According to Yusof *et al.* [27], *Chlorella vulgaris* is a good source of Ca, K, Mg and Zn; these minerals have many functions in human.

Fatty acids composition of oil extracted from *Chlorella vulgaris* biomass are presented in Table 3. As expected the studied oil had elevated amount of total saturated fatty acids (63.24%). This oil had superior content in palmitic acid (C16:0); its percentage was 52.56%, followed by stearic acid (C18:0) (8.83%). It could be noticed that the most predominant unsaturated fatty acids was heptadecanoic (C17:1), its percentage was 15.88%. While, the omega 3 fatty acids linolenic (C18:3) in the second order having a percentage of 8.87 %, followed by linoleic (C18:2) (6.56%) the omega 6 fatty acid, then the mono-unsaturated fatty acid oleic (C18:1) (5.42%). This composition was coincided with the results of Natalia and Nina [28] who found that the most abundant

Table 4: Amino acid composition of *Chlorella vulgaris* biomass protein

Amino acids	Concentration (g/100g protein)
Aspartic acid	10.5
Threonine *	5.24
Serine	5.08
Glutamic acid	10.74
Glycine	5.1
Alanine	8.44
Valine *	6.44
Methionine *	1.5
Isoleucine *	5.01
Leucine *	6.84
Tyrosine	5.2
Phenylalanine*	4.2
Histidin *	5.02
Lysine *	5.6
Arginine	8.2
Proline	6.4
NH4	6.4

*: Essential amino acids

fatty acids were the C16:0 and C18:0, the saturated fatty acids in *Chlorella vulgaris* lipid. The presence of fatty acids with odd number of carbon atoms 17:1 is a firm proof that the algal culture is contaminated with bacteria as mentioned by Petkov and Garcia [29]. It is well known the health benefit of polyunsaturated fatty acids (essential fatty acids), that can be found in plants including microalgae. So, the consumption of n-3 PUFAs from both seafood and plant sources such as Spirulina and *Chlorella* may reduce coronary heart disease (CHD) risk as reported by Mozaffarian *et al.* [30] in a cohort study of 45,722 men. Therefore, *Chlorella* is considered a reliable source of essential fatty acids that are required for many important biochemical functions, including hormone balance.

Chlorella contained about 50 percent of proteins (Table 2), which is rich in essential and nonessential amino acids as shown in Table 4. The presence of nine of essential and seven of non- essential amino acids in the experimented sample of *Chlorella vulgaris* biomass, proof that the cells are capable of synthesizing all amino acids and they can provide the essential ones to human. However, information on the nutritive value of the protein and the degree of availability of amino acids of *Chlorella vulgaris* are given by Janczyk *et al.* [26]. Their overall digestibility is high, which is why there is no limitation for using dried whole *Chlorella vulgaris* microalgae in foods or feeds.

Table 5: Proximate composition (% on dry basis) and penterometer reading (mm) of imitation cheese enhanced with *Chlorella vulgaris* biomass

Properties	<i>Chlorella vulgaris</i> biomass (%)			
	Control	1	2	3
Moisture (%)	39.88 ^a	39.72 ^{ab}	39.28 ^{bc}	38.75 ^c
Protein (%)	20.00 ^a	19.72 ^{ab}	19.41 ^{ab}	19.10 ^b
Fat (%)	30.21 ^a	30.43 ^a	30.54 ^a	30.61 ^a
Salt (%)	5.44 ^a	5.41 ^a	5.40 ^a	5.36 ^a
Ash (%)	4.46 ^a	4.51 ^a	4.66 ^a	4.71 ^a
Fiber (%)	ND	0.15 ^a	0.37 ^{ab}	0.58 ^b
Penterometer reading (mm)	183 ^a	169 ^{ab}	156 ^{bc}	142 ^c

Means with same letters in a row are not significant ($p \leq 0.05$).

ND: Not detected

Table 6: Proximate composition of croissant filled with imitation cheese enhanced with different levels of *Chlorella vulgaris* biomass

Proximate composition (%)	<i>Chlorella vulgaris</i> biomass (%)			
	Control	1	2	3
Moisture	26.30 ^a	25.72 ^a	25.55 ^{ab}	25.08 ^b
Protein	16.55 ^a	16.15 ^a	16.02 ^{ab}	15.43 ^b
Fat	34.61 ^b	34.73 ^b	34.95 ^{ab}	35.75 ^a
Ash	2.12 ^b	2.45 ^{ab}	2.55 ^a	2.76 ^a

Means with same letters in a row are not significant ($p \leq 0.05$).

Physiochemical Properties of Imitation Cheese Enhanced with *Chlorella Vulgaris* Biomass:

As shown in Table 5, the proximate composition of cheese samples in general was slightly and significantly influenced by the addition of the different levels of *Chlorella vulgaris* biomass. Possible, this may be to the small levels of biomass added to the cheese samples, also it could be noticed that, *Chlorella vulgaris* biomass had high percent of total solids, mostly protein and fat. The moisture, protein and salt contents showed a little decrease in cheese samples enhanced with biomass, this reduction increased with increasing the level of *Chlorella vulgaris* biomass to 3%. On contrary, fat, ash and fiber contents increased in the enhanced samples by increasing the level of added biomass (2 and 3%) according to the higher content of these components in the *Chlorella vulgaris* biomass (Table 2). Substitution of rennet casein with *Chlorella vulgaris* biomass produced imitation processed cheeses with the highest firmness values. Accelerated and significant decreases ($P < 0.05$) in penetration value (inverse firmness) were found with the decrease in moisture content of imitation processed cheese samples as illustrated in Table 5. When the moisture was decreased from 39.88 to 38.75%, the decrease in penetration values was from 183 mm to 142mm, respectively. This decrease in the moisture content could be due to the replacement with the biomass which

was higher in the total solids than the rennet casein. The microalgal protein and carbohydrate molecules can also play an important role on the water absorption process, which promotes the increase of cheese firmness, as was observed by Fradique *et al.* [4] and Gouveia *et al.* [31].

Proximate Composition of Croissant Filled with Imitation Cheese Enhanced with Different Levels of *Chlorella Vulgaris* Biomass:

The proximate chemical compositions of croissant samples are summarized in Table 6. Initial observations of croissant samples to which the imitation cheese enhanced with *C. vulgaris* biomass had been added showed that they were slightly different compared to the control. The main difference from the control sample was a decrease in moisture and protein contents, this different was only significant by increasing *C. vulgaris* biomass ratio into the cheese to 3%. Furthermore, fat and ash contents were slightly increased with increasing the ratio of *C. vulgaris* biomass which added to the cheese. The fat of biomass enhanced-croissant ranged between 34.73 and 35.75% being slightly higher than that of the control (34.61%) which was significantly higher ($P < 0.05$) only for the enhanced cheese with 3.0% *C. vulgaris* biomass with higher fat content (Table 5). Microalgae incorporation in the used cheese resulted in an increase of total ash (2.45-2.76%)

content compared to that of the control (2.12%), especially for the croissants prepared with a higher concentration of microalgae biomass. The reasons of the small differences in results were due to the low levels of *C. vulgaris* biomass which added to the used cheese in filling of croissant samples. These results are in agreement with those obtained by Fradique *et al.* [4], who reported that both of moisture and protein contents were decreased while the fat and ash contents were increased in pasta samples enhanced with *C. vulgaris* biomass and the differences were significantly only for the 2.0% enhanced pasta.

Staling Rate (Awrc %) of Croissant Filled with Imitation Cheese Enhanced with Different Levels of *Chlorella Vulgaris* Biomass: Staling is a generic term used to describe the loss of freshness perceived by consumers. It includes crumb firming, development of crumb dryness, loss of flavor and similar changes. The term “staling” refers to the gradually decreasing consumer acceptance of bakery products due to the chemical and physical changes that occur in the crust and crumb during storage, excluding microbial spoilage. Croissant staling were measured and the obtained data are presented in Table 7. It could be noticed that, the filling of imitation cheese enhanced with different levels of *Chlorella vulgaris* did not significantly ($P>0.05$) affected this parameter among all samples and the control from the beginning and during storage. Meanwhile, there was a significant ($P \leq 0.05$)

decrement in staling rate (%) during storage. These results are coincided with those obtained by Baik and Chinachoti [32] who mentioned that the staling process is mainly due to the firming effect caused by moisture transfer from crumb to crust, as well as to the intrinsic firming of the cell wall material, which is associated with starch recrystallization during storage.

Yeasts and Moulds Counts: The yeast and mould counts of different croissant samples are shown in Table 8. It could be seen that, during the three days of storage at room temperature ($18\pm 2^\circ\text{C}$), no moulds or yeasts occurred in any tested samples. Meanwhile, at the sixth day of storage some colonies were appeared ($<5 \log \text{cfu/g}$), this may be due to the efforts made to reduce contamination by using good raw materials and improving hygiene procedures which did not allow the growth of yeast and mould. Data presented in the Table 8 revealed that number of the colonies decreased significantly ($P\leq 0.05$) only in samples of croissant filled with imitation cheese enhanced with 2 and 3% of *Chlorella vulgaris* biomass, this could be due to the antimicrobial effects of algal biomass. The potential antimicrobial properties of algal biomass was mentioned by Queiroz *et al.* [5] who reported that *Chlorella vulgaris* is considered to be a biological response modifier, as demonstrated by its protective activities against viral and bacterial infections in normal and immunosuppressed mice.

Table 7: Staling rate (AWRC %) of croissant filled with imitation cheese enhanced with different levels of *Chlorella vulgaris* biomass during storage at room temperature ($18^\circ \pm 2^\circ\text{C}$)

Storage period (days)	Control	<i>Chlorella vulgaris</i> biomass (%)		
		1	2	3
0	226.6 ^{Aa}	228.2 ^{Aa}	226.6 ^{Aa}	227.7 ^{Aa}
3	217.3 ^{Ab}	219.5 ^{Ab}	218.4 ^{Ab}	215.6 ^{Ab}
6	197.1 ^{Ac}	198.2 ^{Ac}	194.1 ^{Ac}	195.0 ^{Ac}

Means followed by the same capital letters in the same row; and values followed by the same small letters in the same columns are not significantly different ($P \leq 0.05$).

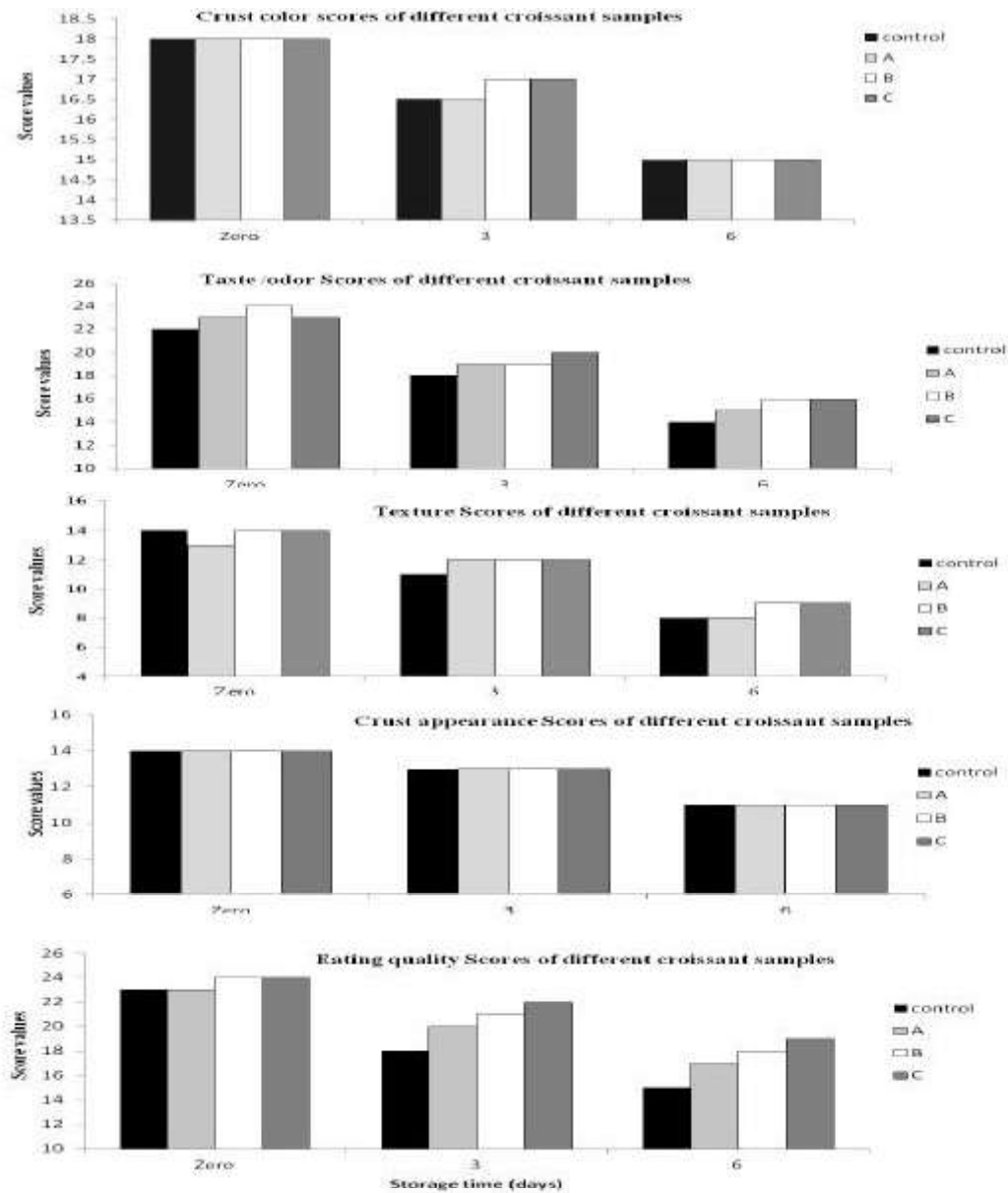
Table 8: Yeast and mould counts ($\log \text{cfu} / \text{g}$) of croissant filled with imitation cheese enhanced with different levels of *Chlorella vulgaris* biomass during storage at room temperature ($18 \pm 2^\circ\text{C}$).

Storage period (days)	Control	<i>Chlorella vulgaris</i> biomass (%)		
		1	2	3
0	ND	ND	ND	ND
3	ND	ND	ND	ND
6	2.60 ^a	2.34 ^a	1.91 ^b	1.60 ^b

Cfu: colony forming unit

ND: Not detected

Means with same letters in a row are not significant ($P \leq 0.05$).



Control: croissant filled with imitation cheese

A: croissant filled with imitation cheese enhanced with 1 % *Chlorella vulgaris*

B: croissant filled with imitation cheese enhanced with 2 % *Chlorella vulgaris*

C: croissant filled with imitation cheese enhanced with 3 % *Chlorella vulgaris*

Fig. 1: Sensory evaluation of croissant filled with imitation cheese enhanced with different levels of *Chlorella vulgaris* biomass during storage at room temperature ($18 \pm 2^\circ\text{C}$).

Sensory Evaluation of Croissant Filled with Imitation Cheese Enhanced with Different Levels of *Chlorella Vulgaris* Biomass: Changes in the sensory attributes of the four kinds of croissant samples during storage periods (days) at $18 \pm 2^\circ\text{C}$ are shown in Fig. 1. There were no significant differences ($P > 0.05$) in crust color and

appearance among the different samples and during storage period, this was due to the non influence of the used filling contained imitation cheese on these parameters. It could be notice that, with increasing the amount of replacing rennet casein with *Chlorella vulgaris* biomass in imitation cheese that used in filling the

croissant, the taste and odor were enhanced and were given high scores by panelists. At zero time of storage, there were significant differences ($P<0.05$) between samples in both parameters, the highest scores were appeared in croissant samples which filled with imitation cheese enhanced with 2 % *Chlorella vulgaris* biomass followed by those filled with 3% *Chlorella vulgaris* biomass. As storage period was elongated, there were significant ($P<0.05$) deterioration between samples, meanwhile, the samples which filled with *Chlorella vulgaris* biomass had higher scores when compared to control one which indicated that the addition of *Chlorella vulgaris* biomass enhanced taste / odor in croissant samples during storage as mentioned by Yamaguchi [33] who reported that *Chlorella* can also be used as a food additive owing to the taste and flavor-adjusting actions of its coloring agent. From the same data, it could be noticed that, the texture of control sample received significantly ($P<0.05$) lower texture score values than other croissant samples during the storage period. Both 2 and 3 % of *Chlorella vulgaris* biomass in imitation cheese showed a significant ($P<0.05$) influence on this parameter. It could be also noticed that, there were significant ($P<0.05$) differences between samples in their eating quality from the beginning and during storage, this was due to the filling material which enhanced by microalgae. With increasing the amount of replacing of rennet casein with *Chlorella vulgaris* biomass, used in filling the croissant, the eating quality scores showed noticeable increase as compared to the control sample. These results are in agreement with those reported by Fradique *et al.* [4] who found that pastas prepared with 2% of green *Chlorella vulgaris* was better for the panelists. Results of the sensory analysis of croissants filled with imitation cheese enhanced with different quantity of *Chlorella vulgaris* biomass showed that using of this microalgae in imitation cheese, could improve the sensory attributes of croissants. The levels of 2 and 3 % of this microalga showed significant impact on better croissants taste, texture and eating quality. These results are in accordance with the aim of this work to develop innovative croissant that are attractive to consumers with similar texture and flavor profiles to the traditional common croissant.

CONCLUSIONS

The abundance of bioactive components in *Chlorella vulgaris* is of great importance from a nutritional point of view because thus the *Chlorella*

vulgaris biomass provides a new opportunity for the manufacture of functional foods. Croissant, a traditional and nutritious food, can be healthy and very attractive when prepared with the addition of a natural microalgal biomass of *Chlorella vulgaris* to the cheese which used as a filling agent in croissant. The addition of *Chlorella vulgaris* biomass to croissants may be an appealing way to increase the daily intake of functional nutrients, especially antioxidants and polyunsaturated fatty acids, essential amino acids and vitamins, which were very important to health promotion and prevention of many kinds of disorders such as gastric ulcers, constipation, anemia, hypertension, diabetes, infant malnutrition and neurosis. The addition of biomass as natural ingredient resulted in croissants with an attractive and innovative appearance and higher textural characteristics. *Chlorella vulgaris* croissant presented good profile tonalities, so the obtained croissant reveals a new niche food market.

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