Quality Evaluation of Mayonnaise Enriched with Ethanolic Chard Leaves Extract

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Quality Evaluation of Mayonnaise Enriched with Ethanolic Chard Leaves Extract

*Azza S. Abdel-Ghany** *Ghada M. El-Araby***

Abstract:

Plant extracts can have a significant role in the shelf-life prolongation of full-fat foods. So, the main objectives of the present study were to assess the effect of ethanolic chard leaves extract addition on the physicochemical, sensorial and microbiological mayonnaise properties. The ethanolic chard leaves extract was added to mayonnaise with levels 0.5. 1.0 and 1.5% of mayonnaise weight. Then, these samples were compared with mayonnaise samples fortified with tert-butyl hydroquinone (TBHQ) as the artificial antioxidant and with a sample without antioxidants as the control. The results displayed that the chard leaves had a high content of protein, ash and total phenolic content. The radical scavenging activity of ethanolic chard leaves extract was 90.11±1.21% compared to 95.22±0.87% for TBHQ at 120 min. An addition of chard leaves extract did not affect the chemical composition of mayonnaise. But it led to a shiny beige color and lost part of the polish appearance of samples. The mayonnaise containing 1.5% chard leaves extract had a non-significant decrease in most of the sensory evaluation scores, except the color, compared with the mayonnaise containing TBHQ and control. No significant differences in TBA values were observed between mayonnaise samples containing TBHQ and 1.5% chard leaves extract at different storing periods. Moreover, the TBA values, titratable acidity, the total bacterial, yeast and molds count increased in all mayonnaise treatments by progressing storage periods. Inversely, the viscosity and pH values decreased in all treatments by progressing storage periods. It can be recommended that the potentiality of using the extract of the chard leaves as a natural antioxidant during mayonnaise manufacture to

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delay oxidation and prolong the shelf-life of mayonnaise instead of chemical antioxidants

Keywords: Mayonnaise, antioxidant, chard leaves extract, physico-chemical properties

Introduction

Mayonnaise is a type of the most popular dressing species in the world. It is a semi-solid, oil-in-water, emulsion which is produced with good mixing of egg yolk, vinegar, oil, salt and spices as mustard and black pepper) (**Mirzanajafi-Zanjani *et al.* 2019)**. Egg yolk is a significant factor during mayonnaise processing because of its high emulsifying ability, which binds to phospholipids, lipoproteins, and unbound proteins, thus maintaining the mayonnaise stability **(Ghazaei *et al*. 2015).**

Fat is one of the base ingredients in mayonnaise as it is represented 70%–80%. It has a significant relation with the rheological, sensory and technical properties especially the texture of the final product. The manufacture of low-fat mayonnaise is a negative effect on some technical properties like stability, texture, mouth feeling and flavor. Mayonnaise is a high oil-containing food, so it is squeamish to oxidation, thus leading to its quality decadence and undesirable compounds formation such as aldehydes and free radicals **(Amin *et al*., 2014)**.

Mayonnaise is a fatty food which is susceptible to deterioration **owing to** the oxidation of the unsaturated fatty acid in oil **(Abu-Salem and Abou-Arab, 2008)**. Lipid oxidation rate in the mayonnaise is affected by many different factors as the oil type used the structure molecular fats, light, heat, physical features of emulsion droplets, and manufacturing environments. Also, the low pH of mayonnaise (pH 4) reasons flouting of the iron bridges among egg yolk proteins and freeing of the iron which can *contribute* in the oxidation of unsaturated fatty acids, making lipid radicals or cause degradation of peroxides **(Romeo *et al*., 2021)**.

Synthetic antioxidants have been used to delay or prevent the lipid oxidation reactions and increase the stability of oxidation in foods, but using them for a long time cause the possible potential toxicity and some serious

diseases appearance **(Lachance *et al*., 2001)**. In recent years, natural antioxidants have gained prominence as alternatives to synthetic antioxidants. The natural antioxidants are existing at different levels in plants for example fruits, vegetable leaves, flowers, roots, grains and seeds. Consequently, the natural extracts have been output and used in the food industries to block of fat oxidation and prolong the storage period of fatty foods **(Etti *et al*., 2013).** Moreover, **Radford and Board (1993)** exposed that the existence of some vegetable substances in the mayonnaise has the consequence of blocking bacteria development and reducing mayonnaise poisoning.

Chard (*Beta vulgaris L. var. cicla*) is one of the healthiest green leafy vegetables which are considered a good source of many nutrients and phytochemicals. Chard contains significant concentrations of nutritional factors such as polysaccharides, vitamins (A, C, B and folic acid), minerals (calcium, iron and phosphorus) and some greasy acids (stearic, oleic, palmitic, linoleic and linolenic acids) **(Zein *et al*., 2015)**. Its stems are high in potassium content, but the leaves had the highest in fiber, magnesium, sodium, vitamin C and flavonoids content **(Gamba *et al*., 2020)**.

Chard has multiple healthy benefits as decreasing the risk of obesity, diabetes, heart disease, reducing blood pressure, improving endothelial dysfunction and inhibiting platelet aggregation. Chard leaves have many activities as antioxidant, anticancer, hepatoprotective, nephroprotective, anti-inflammatory and wound healing **(Hajihosseini *et al*., 2017)**. Also, it contains chlorophyll and betalains, which is efficacious to retardant the heterocyclic amines caused cancer, that is formed by high heat during grilling foods **(Kugler *et al*., 2004 and Lidder & Webb, 2013).**

The phenolic compounds are potent antioxidants and free radical scavengers. The antioxidant activity of phenolics is mainly due to their redox properties, which allow them to act as reducing agents, hydrogen donors and singlet oxygen quenchers **(Zein *et al*., 2015)**. A significant content of phenolic compounds and flavanol glycosides with relevant high antioxidant activity are present in chard. Some studies suggest that chard

provides a good natural source of antioxidant and anti-acetylcholinesterase activities **(Sacan and Yanardag, 2010).**

Therefore, this study was aimed to assess the antioxidant activity of ethanolic chard leaves extract and its effect as a natural antioxidant on chemical, physical, sensorial and microbiological properties of mayonnaise during refrigerated storing at 4°C.

Materials and methods

Materials

The chard plant was purchased from the local market at Zagazige city, Sharkia Governorate, Egypt. Chard leaves were separated manually, washed with distilled water, and dried at room temperature and completely dried at air oven (40±2˚C). The dried leaves were grind by a café mill and was sieved through a 60mesh screen and stored in a tightly sealed plastic container in the freezer at - 18° C for further uses and analysis.

Preparation of ethanolic chard leaves extract

Dried powder of chard leaves was extracted in the laboratory using aqueous ethanol 70% at ratio (1:10 w/v), overnight at room temperature with shaking, followed by filtration through Whatman paper (No.1). The residues were re-extracted under the same conditions then the combined filtrate was evaporated in a rotary evaporator (BÜCHI-water bath-B-480, Germany) at 45°C. The extract was freeze-dried with freeze dryer model Thermo- Electron Corporation, Heto power dry LL300, France. The extract was weighed to determine the yield and stored at -20 °C until further use, according to **Contini *et al*. (2008).**

Determination of total phenolics and flavonoids for extract

The total phenolic was measured by using a Folin–Ciocalteu reagent as described by **Singleton and Rossi (1965)**, the phenols were measured at 765nm then the results were reported as mg of gallic acid equivalents (GAE) per g of chard leaves extract. Total flavonoid content was determined as **Ordon *et al*. (2006)**, the absorbance was measured at 420 nm, and

flavonoids content was expressed as mg quercetin equivalent (QE) per g of chard leaves extract.

Antioxidants activity assay

The free radical scavenging of ethanolic extract was measured by the 2,2- diphenyl-1 hydrazyl (DPPH) assay according to **(Burits and Bucar, 2000)** with some modification, briefly 3ml of 0.1mM ethanolic solution of DPPH was added at concentration 100 µg/ml to 1 ml of ethanolic extract. The absorbance was measured against a blank at 517 nm at 0, 30, 60, 90 and 120 min. Inhibition of free radical DPPH in percent was calculated by the following equation:

$$\text{DPPH scavenging activity\%} = (Ac- As/ Ac) \times 100;$$

Where Ac is the absorbance of the control reaction (containing all reagents except the extract) and as is the absorbance in the presence of the tested extract.

Preparation of mayonnaise

Mayonnaise samples were prepared according to **Kishk and Elsheshetawy (2013)** using the formula contained the following ingredients: sunflower oil 700g, whole egg 191g, salt 10g, sugar 6g, lemon juice 16g, vinegar 56g, mustard 18g, and white pepper 3g. The preparation was carried out by mixing egg, vinegar, and then other ingredients using an electric mixer (Bimby TM31, Vorwerk, Wuppertal, Germany) for 5 sec. Then, the oil was slowly added under a vigorous mixing rate (from 3200 rpm up to 6000 rpm in 5 min). The resultant mayonnaise was divided into five equal portions and treated as follow; the first portion was retained without additives and served as a control (C), the second portion was treated with TBHQ as a synthetic antioxidant at a level of 200ppm (T1), The third portion was treated by ethanolic chard leaves extract at a concentration of 0.5% (T2), the fourth portion treated by 1.0% of chard leaves extract (T3) and the fifth portion treated with 1.5% of chard leaves extract (T4). The prepared mayonnaise samples were filled in covered cup (100 g size) and kept at 4ºC for 21 days. All mayonnaise samples were examined chemically,

microbiologically and organoleptically at the1st, 7th, 14th and 21 days of the storing.

Chemical analysis

Moisture, ash, crude fat and crude protein of chard leaves powder and mayonnaise samples were calculated by multiplying total nitrogen value by a factor of (6.25) according to **AOAC (2005).** Total carbohydrate contents were estimated by difference = 100- (moisture% + crude oil% + crude protein% + ash %). The pH of mayonnaise samples was measured at 4ºC by a pocket pH meter (Model IQ 125, IQ Scientific USA). Titratable acidity of mayonnaise samples was determined (**AOAC, 2000).**

Determination of thiobarbituric (TBA) value

TBA values of mayonnaise samples were detected according to the methods of **Keeny (1971).**

Rheological properties

The mayonnaise viscosity was measured according to **Liu *et al.* (2007)** by a rotational viscometer (model 5437). The samples color was measured by Hunter lab color analyzer (Hunterlab Colour Flex EZ, USA).

Sensory evaluation

The acceptability of mayonnaise product was defined by sensory evaluation of all treatments according to **Kishk and Elsheshetawy (2013)** by 12 panelists from the memberships of the Food Science Department, Faculty of Agriculture, Zagazig University.

Microbiological tests

All mayonnaise samples were enumerated for total bacterial count *Staphylococcus aureus* and *Salmonella spp*. according to **American Public Health Association (1992)**. Yeasts and molds were counted by **Difco (1984)** method on acidified potato dextrose agar medium.

Statistical analysis

Statistical analyses of all data were performed by Statistix 8.1 Package Program **(Statistix, 2009)**. Mean ± SD was used to describe all

data. Least significant difference (LSD) test was used to found the significant differences between all treatments and the storage periods.

Results and discussion

Proximate chemical composition of chard leaves powder

The chard leaves chemical composition is detailed in Table (1). The moisture content to chard leaves powder was 6.82± 0.77%. Total protein, fat, ash and carbohydrate content were 25.27± 2.01, 1.12± 0.02, 17.75± 1.13 and 49.04±3.86%, respectively. Moreover, the caloric values of chard leaves were 307.31± 23.66 Kcal/100g. It is observed from the results that chard leaves had a high content of protein and ash content. **Mzoughi *et al*. (2019)** found that the moisture, fat, protein and ash contents in fresh chard leaves were 93.35±0.29, 0.099±0.01, 0.663±0.01 and 1.30±0.06 g/100 g fresh weight.

Table (1): Chemical composition of chard leaves

Component	Values
Moisture (%)	6.82± 0.77
Crude fat (%)	1.12± 0.02
Crude protein (%)	25.27± 2.01
Ash (%)	17.75± 1.13
Carbohydrate (%)	49.04±3.86
Caloric values (Kcal/100g)	307.31± 23.66

Total phenolics and flavonoids content

The chard leaves extract content of total phenolics and flavonoids were 101.95± 3.44 mg GAE/g extract and 51.71± 1.64 mg QE/g extract, respectively (Table 2). The total phenolics and flavonoids content in our study are higher than that found by **Hamdiken and Kechrid (2017)**, who found that chard extract contained 31.23±0.007 mg GAE and 6.85±0.087 mg QE /g extract, respectively. Also, **Mzoughi *et al*. (2019)** stated that the content of phenolic and flavonoid compounds in chard leaves extract were 96.58 ±1.81 mg GAE and 30.08 ± 1.02 mg Catechin E /g extract,

respectively. **Pyo *et al.* (2004)** mentioned that the chard is a good dietary source of phenolic compounds.

Table (2): Total phenolics and flavonoids content of chard leaves extract

Component	Values
Total phenolics (mg GAE/g extract)	101.95± 3.44
Flavonoids (mg QE/g extract)	51.71± 1.64

Antioxidant activity

The DPPH radical scavenging activity of ethanolic chard leaves extract compared with TBHQ are denoted in Fig. (1). The results indicated that the radical scavenging activity was 90.11±1.21% compared to 95.22±0.87% for TBHQ at 120 min. The results suggest that chard leaves extract is capable of scavenging free radicals by their phenolic structure which act as hydrogen or electron donor. Where, the phenoxy radicals formed in the reaction of antioxidant with fatty acid peroxy radical is steadied by delocalization of the impaired electrons around the aromatic ring **(Ramadan *et al*., 2003).** Where **Pyo *et al*. (2004) and Zein *et al*. (2015)** indicated that there was a significant linear correlation of the total phenolic compounds level with the DPPH scavenging activity to chard extracts. Significant differences were found between chard leaves and stems in the antioxidant activity.

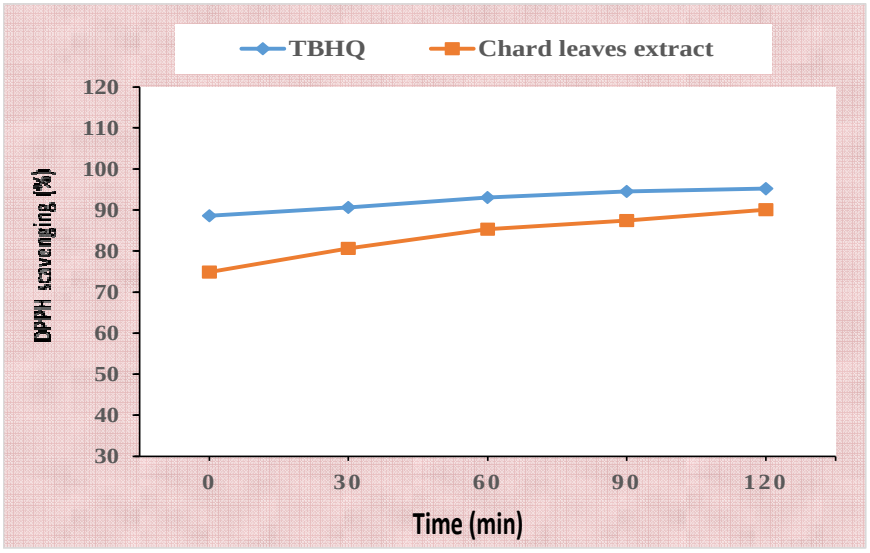

Fig. (1): DPPH scavenging activity of ethanolic chard leaves extract and TBHQ

Chemical composition of mayonnaise

The chemical composition of mayonnaise produced in this study is shown in Table (3). No significant differences were found among all mayonnaise treatments and control samples to all chemical composition parameters and caloric values. The high caloric values of all mayonnaise samples were a result of increased fat used in processing. In comparison with **USDA (2014)** data which reported that the full-fat mayonnaise is contained an energy content of approximately 700-800 kcal, our results recorded lower energy content in mayonnaise. **Marinescu *et al*. (2011) and Al-Aubadi (2021**) indicated it can be overcome that by using some suitable fat replacers such as β- glucan, chitosan and tamarind gum to produce healthy low-fat mayonnaise without any change in the properties of the final product.

Table (3): Chemical composition (%) of mayonnaise samples

Treatments	C	T1	T2	T3	T4	LSD
Moisture	24.34±1.46[a]	23.51±1.18[a]	23.98±2.76[a]	24.01±0.42[a]	23.61±1.09[a]	2.88
Crude fat	70.68±0.88[a]	70.92±0.70[a]	70.58±0.52[a]	70.82±0.64[a]	70.99±0.36[a]	1.17
Crude protein	1.06±0.32[a]	1.21±0.11[a]	1.17±0.23[a]	1.04±0.31[a]	1.18±0.05[a]	0.42
Ash	0.87±0.24[a]	0.91±0.21[a]	0.92±0.17[a]	0.81±0.10[a]	0.86±0.29[a]	0.38
Carbohydrate	3.05±1.40[a]	3.45±1.69[a]	3.35±2.38[a]	3.32±0.47[a]	3.36±1.17[a]	2.83
Caloric values (Kcal/100g)	652.59±10.39[a]	656.92±3.46[a]	653.30±13.64[a]	654.79±5.12[a]	657.06±6.72[a]	15.81

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same row.

Sensory evaluation

The sensory evaluation results of the prepared mayonnaise samples are presented in Table (4). The mayonnaise control sample had the highest scores for all the sensory attributes followed by mayonnaise containing TBHQ. The appearance of the control sample was homomorphic, bushy and furbished but the mayonnaise containing chard leaves extract lost part of the polish appearance.

A significant difference was found in the color score between mayonnaise containing 0.5, 1.0 and 1.5% chard leaves extract and the control. The C and T1 samples had a glossy yellow color whilst the samples containing chard leaves extract had a shiny beige color. This is due to the incorporation of chard leaves extract caused to change color. The changing color increased with increasing the chard leaves extract ratio in mayonnaise, hence was reflected on L^* values (Table 5). The flavor was not significantly influenced by the chard leaves extract addition. The control had the highest consistence compared to other mayonnaise treatments and the T3 sample had the lowest consistence.

As regards the overall acceptability of mayonnaise, it can be observed from the table that the control and T1 samples had the highest acceptability and the T3 had the lowest acceptability. Also, it could be noted

that T3 recorded the lowest scores in most of the sensory attributes. While T4 had a non-significant decrease in most of the sensory evaluation scores compared with the control except the color.

Table (4): Sensory evaluation of mayonnaise samples

Treatments	C	T1	T2	T3	T4	LSD
Appearance	8.13 ± 0.93^{a}	7.58 ± 0.90^{abc}	7.21 ± 1.05^{bc}	7.00 ± 0.74^{c}	7.92 ± 0.67^{ab}	0.71
Taste	8.00 ± 0.85^{a}	7.17 ± 0.94^{bc}	7.00 ± 0.95^{c}	6.75 ± 1.14^{c}	7.83 ± 0.83^{ab}	0.78
Flavor	7.33 ± 1.07^{a}	7.00 ± 1.35^{a}	6.96 ± 1.14^{a}	7.50 ± 1.38^{a}	7.92 ± 1.08^{a}	0.99
Color	8.58 ± 0.47^{a}	8.50 ± 0.52^{a}	7.68 ± 0.56^{b}	7.05 ± 0.99^{bc}	6.79 ± 0.81^{c}	0.60
Consistency	7.67 ± 0.78^{a}	7.04 ± 1.16^{ab}	7.17 ± 1.11^{ab}	6.50 ± 1.09^{b}	7.50 ± 1.17^{a}	0.88
Mouthfeel	7.92 ± 1.00^{a}	7.46 ± 0.75^{ab}	6.92 ± 1.31^{bc}	6.46 ± 1.41^{c}	7.38 ± 1.37^{abc}	0.98
Overall acceptability	8.42 ± 0.67^{a}	8.25 ± 0.45^{a}	7.50 ± 0.88^{b}	6.42 ± 0.79^{c}	8.00 ± 0.95^{ab}	0.63

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same row.

Color characteristics:

Color characteristics of mayonnaise samples are illustrated in Table (5). The mayonnaise color changed by the addition of chard leaves extract. No significant variances were noted in color values between mayonnaise containing TBHQ and the control sample. Lightness values (L^*) of mayonnaise containing chard leaves extract were significantly lower than the control samples. Moreover, during storage periods at 4°C for 21 days, the lightness values during storage for 7 days of all mayonnaise samples were significantly higher than the other storage periods.

Conversely, the redness (a^*) values were high significant in all mayonnaise treatments compared to the control sample. Concerning yellowness (b^*) values, mayonnaise containing 1.0 and 1.5% chard leaves extract were significantly higher than the other treatments and control samples. There were no significant differences in a^* and b^* values during all storage periods of mayonnaise treatments except b^* values of T1 on 21 days and T2 on 1st day. These results are in accordance with

(Chotphruethipong and Benjakul, 2018), who found that the addition of cashew leaves extract to mayonnaise with 100 and 200 mg/kg increased *a** and *b** but decreased *L** values.

Table (5): Color characteristics of mayonnaise samples during storing at 4°C for 21 days

Treatments	Storing periods (days)	C	T1	T2	T3	T4	LSD
*L**	1	$85.33\pm0.02^{A,c}$	$85.50\pm0.38^{A,a}$	$78.38\pm0.05^{B,b}$	$72.32\pm0.07^{C,b}$	$69.31\pm0.06^{D,c}$	0.32
	7	$86.21\pm0.02^{A,a}$	$86.02\pm0.17^{A,a}$	$78.88\pm0.04^{B,a}$	$73.68\pm0.30^{C,a}$	$70.79\pm0.16^{D,a}$	0.31
	14	$84.65\pm0.09^{B,d}$	$85.62\pm0.98^{A,a}$	$78.10\pm0.12^{C,c}$	$71.91\pm0.05^{D,c}$	$70.21\pm0.38^{E,b}$	0.87
	21	$85.51\pm0.05^{A,b}$	$85.39\pm0.05^{A,a}$	$78.44\pm0.19^{B,b}$	$72.60\pm0.07^{C,b}$	$70.33\pm0.25^{D,ab}$	0.27
LSD		0.10	1.00	0.22	0.30	0.46	
*a**	1	$-2.07\pm0.03^{A,a}$	$-2.28\pm0.25^{A,a}$	$-3.14\pm0.06^{B,a}$	$-3.85\pm0.15^{C,a}$	$-4.02\pm0.04^{C,a}$	0.24
	7	$-2.14\pm0.08^{A,a}$	$-2.11\pm0.03^{A,a}$	$-3.30\pm0.27^{B,a}$	$-4.07\pm0.97^{BC,a}$	$-4.12\pm0.03^{C,a}$	0.82
	14	$-2.15\pm0.13^{A,a}$	$-2.27\pm0.05^{A,a}$	$-3.42\pm0.09^{B,a}$	$-4.17\pm0.04^{C,a}$	$-4.20\pm0.20^{C,a}$	0.21
	21	$-2.06\pm0.04^{A,a}$	$-2.12\pm0.11^{A,a}$	$-3.28\pm0.25^{B,a}$	$-4.05\pm0.05^{C,a}$	$-4.18\pm0.02^{C,a}$	0.23
LSD		0.15	0.26	0.36	0.92	0.19	
*b**	1	$15.58\pm0.55^{B,a}$	$15.77\pm0.19^{B,a}$	$16.61\pm0.06^{B,b}$	$19.09\pm1.07^{A,a}$	$19.72\pm0.99^{A,a}$	1.27
	7	$15.93\pm1.03^{C,a}$	$15.85\pm0.11^{C,a}$	$17.18\pm0.03^{B,ab}$	$19.10\pm0.85^{A,a}$	$19.63\pm0.04^{A,a}$	1.09
	14	$15.91\pm0.96^{C,a}$	$15.31\pm0.24^{C,a}$	$17.47\pm0.38^{B,a}$	$19.69\pm0.05^{A,a}$	$19.99\pm1.00^{A,a}$	1.18
	21	$14.75\pm1.09^{C,a}$	$14.60\pm0.65^{C,b}$	$17.40\pm0.54^{B,a}$	$18.96\pm0.95^{A,a}$	$19.21\pm0.06^{A,a}$	1.36
LSD		1.75	0.68	0.62	1.57	1.32	

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively

*L**: lightness, *a**: redness, *b**: yellowness.

The superscript capital letters are differed significantly ($P \leq 0.05$) in the same row.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same column.

Thiobarbituric acid (TBA) value

TBA value is an index to assess the development of oxidation changes that appeared in fatty foods. It determines the levels of secondary

oxidation products like aldehydes, malonaldehyde and ketones **(Farag *et al*., 1990)**. The changes in the TBA of mayonnaise treatments are presented in Table (6). The TBA values of mayonnaise containing TBHQ and ethanolic chard leaves extract were lower than in the control sample. There was no significant difference between mayonnaise samples containing TBHQ and 1.5% chard leaves extract at different storage periods in TBA values. This is due to phenolic compounds found in extracts, where it acts as hydrogen or electron donors to proxy radicals in the reaction and thus decreasing the formation of the hydroperoxides and secondary products or delaying the autoxidation of unsaturated fatty acid **(Rasmy *et al*., 2012).**

Moreover, the TBA values increased in all treatments by progressing storage periods. This increase was significant in the control and mayonnaise samples containing 0.5 and 1.0% chard leaves extract and insignificant in mayonnaise containing TBHQ and 1.5% chard leaves extract. These results match with **El-Rahman *et al.* (2020),** who found that the TBA values increased for the control and samples treated with BHA and moringa leaves extract by the ending of the storage time.

Table (6): TBA values (mg malonaldehyde/kg fat) of mayonnaise samples during storing at 4°C for 21 days

Storing periods (days)	C	T1	T2	T3	T4	LSD
1	$0.228\pm0.025^{A,b}$	$0.202\pm0.003^{A,a}$	$0.218\pm0.018^{A,c}$	$0.212\pm0.013^{A,b}$	$0.210\pm0.020^{A,a}$	0.032
7	$0.245\pm0.007^{A,b}$	$0.208\pm0.006^{C,a}$	$0.241\pm0.006^{AB,bc}$	$0.231\pm0.003^{BC,ab}$	$0.223\pm0.011^{C,a}$	0.012
14	$0.286\pm0.025^{A,a}$	$0.212\pm0.014^{C,a}$	$0.261\pm0.001^{AB,ab}$	$0.251\pm0.022^{B,a}$	$0.230\pm0.021^{BC,a}$	0.033
21	$0.299\pm0.012^{A,a}$	$0.219\pm0.017^{C,a}$	$0.285\pm0.021^{A,a}$	$0.271\pm0.031^{AB,a}$	$0.239\pm0.028^{BC,a}$	0.040
LSD	0.036	0.021	0.026	0.038	0.039	

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript capital letters are differed significantly ($P \leq 0.05$) in the same row.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same column.

The mayonnaise viscosity

Fig. (2) display the mayonnaise samples viscosity within storing at 4°C during 21 days. It was obvious that the viscosity values of the mayonnaise samples containing 1.0 and 1.5% chard leaves extract were greater than other treatments during 1st, 7th and 14 days of storage periods. The lowest value in viscosity was observed for samples containing 1.0 % chard leaves extract on 21 day of storage periods. Also, the viscosity values significantly decreased in all treatments by progressing storing periods. These results are harmony with **(Abu-Salem and Abou -Arab, 2008).**

The mayonnaise viscosity and stability are high when the pH is thereabout to the isoelectric point of the egg yolk proteins, thus the charged proteins are reduced. If the proteins on the droplets surface were fully charged, this would block any extra protein from absorbing and led to indispose the droplets about one to another, which would block flocculation, thus leading to low viscosity and stability **(Depree & Savage, 2001 and Mirzanajafi-Zanjani *et al.*, 2019).** Therefore, the mayonnaise viscosity decreased during the storage period as a result of a decrease in the mayonnaise pH **(Triawati *et al.*, 2016).**

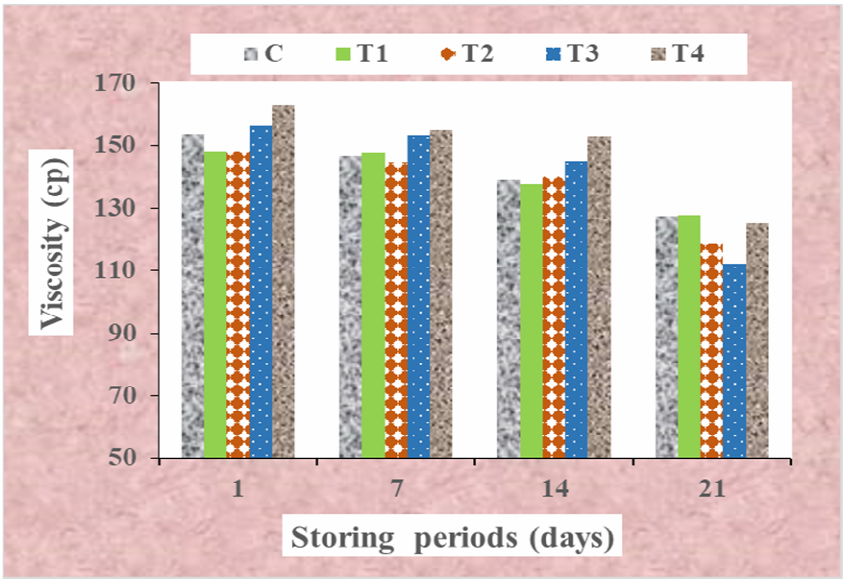

Fig. (2): Viscosity values changes of mayonnaise samples within storing at 4°C during 21 days

pH values

The mayonnaise pH values are illustrated in Table (7). No significant differences were noted in pH values among the control and the other mayonnaise treatments during the different storing periods. On the other side, the pH values of all treatments are reduced with progressing storing periods. The lowest reduction in pH values was observed for mayonnaise samples containing 1.0% chard leaves extract followed by T1 and T4 on 21 days of storing.

Reducing of the mayonnaise pH during storing due to the growth of microorganism, which produces more organic acids. The high content of organic acids decreases the mayonnaise pH values thence caused some changes in the physical properties of mayonnaise **(Triawati *et al*., 2016).** These results are in accordance with **(Abu-Salem & Abou -Arab, 2008 and Nour, 2021).** In contrast, **Rasmy *et al*. (2012)** mentioned that pH values increased during storing periods for the samples of mayonnaise control, containing BHA and sage extract with different concentrations.

Table (7): The mayonnaise pH values during storing at 4°C for 21 days

Storing periods (days)	C	T1	T2	T3	T4	LSD
1	$4.37\pm0.19^{A,a}$	$4.39\pm0.28^{A,a}$	$4.42\pm0.23^{A,a}$	$4.41\pm0.04^{A,a}$	$4.31\pm0.17^{A,a}$	0.36
7	$4.30\pm0.21^{A,a}$	$4.26\pm0.16^{A,ab}$	$4.37\pm0.06^{A,a}$	$4.35\pm0.27^{A,a}$	$4.21\pm0.08^{A,ab}$	0.32
14	$4.19\pm0.18^{A,a}$	$4.20\pm0.09^{A,ab}$	$4.28\pm0.25^{A,a}$	$4.26\pm0.22^{A,ab}$	$4.16\pm0.04^{A,ab}$	0.31
21	$3.98\pm0.25^{A,a}$	$4.02\pm0.19^{A,b}$	$4.09\pm0.16^{A,a}$	$3.94\pm0.24^{A,b}$	$4.05\pm0.08^{A,b}$	0.35
LSD	0.39	0.36	0.36	0.40	0.20	

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript capital letters are differed significantly ($P \leq 0.05$) in the same row.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same column.

Titratable acidity

Table (8) illustrates the changes in titratable acidity% of mayonnaise samples during storing at 4°C to 21 days. The results showed that, the

titratable acidity% of mayonnaise containing TBHQ and ethanolic chard leaves extract were lower significant than that of the control samples during all the storing periods. Moreover, the titratable acidity% of all mayonnaise treatments significantly increased on 21 days compared with 1st day of storage periods. During storage, the growth of some bacterial groups like lactic acid bacteria led to an increase the acidity, hence, pH values were decreased **(Worrasinchai *et al*., 2006).**

Table (8): Titratable acidity of mayonnaise samples during storing at 4°C for 21 days

Storing periods (days)	C	T1	T2	T3	T4	LSD
1	$0.387\pm0.015^{A,b}$	$0.200\pm0.030^{D,c}$	$0.320\pm0.020^{B,b}$	$0.287\pm0.040^{BC,b}$	$0.250\pm0.010^{C,b}$	0.05
7	$0.430\pm0.036^{A,ab}$	$0.217\pm0.021^{C,bc}$	$0.326\pm0.013^{B,b}$	$0.313\pm0.006^{B,ab}$	$0.287\pm0.085^{BC,ab}$	0.08
14	$0.460\pm0.060^{A,a}$	$0.263\pm0.042^{C,ab}$	$0.357\pm0.006^{B,a}$	$0.317\pm0.015^{BC,ab}$	$0.315\pm0.013^{BC,ab}$	0.06
21	$0.484\pm0.012^{A,a}$	$0.290\pm0.026^{C,a}$	$0.379\pm0.010^{B,a}$	$0.347\pm0.045^{B,a}$	$0.341\pm0.011^{B,a}$	0.05
LSD	0.07	0.06	0.02	0.06	0.08	

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript capital letters are differed significantly ($P \leq 0.05$) in the same row.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same column.

Microbiological analysis

The results in Table (9) illustrate the total bacterial, *Salmonella spp*, *Staphylococcus aureus* and yeasts and molds counts of all mayonnaise treatments during storing at 4°C to 21 days. The total bacterial count of mayonnaise samples containing TBHQ and chard leaves extract was significantly lower than the control through all the storing periods, except T2 and T3 on 14th day of storage. Also, the total bacterial count significantly increased with progressing storing periods for the mayonnaise control and treatments. The reduction of total bacterial counts on 1st day of storing may be due to the impact of undissociated and soluble acetic acid into the oil phase. After storing period, the total count in the control increased probably

by reason of acid tolerant micro-organisms development **(Marinescu *et al*., 2011).**

The yeasts and molds were not detected on the 1st day of the storing period for the control and all the other treatments. Also, it was not discovered on the 7th day for T4. The yeast and molds count of the mayonnaise control and treatments were discovered on the 7th day of storing and then increased till the end of the storing period. The lowest counts of yeasts and molds were observed on the 7th and 14th days of storage for T1 and the 7th days for T3.

Meanwhile, the *Salmonella spp* and *Staphylococcus aureus* bacteria were not discovered in the control and other mayonnaise treatments during all storing periods. Home-prepared mayonnaise has been related to outbreaks of food poisoning resulting to the presence of *Salmonella* bacteria which due to eggs are the main source of infection with it. The results of this study found that *Salmonella spp* and *Staphylococcus aureus* bacteria not detected in any treatments, this may be due to the use of vinegar (acetic acid), lemon juice (citric acid) and mustard which is acted a germicidal and increased in the rate of bacteria death. Also, **Xiong *et al*. (2000)** stated that the added vinegar within mayonnaise processing (6% acetic acid) is desired to produce Salmonella-free mayonnaise in the kitchen. Moreover, the storing of mayonnaise in cold temperatures prevents bacteria growth. The presence of natural extracts in mayonnaise is an antimicrobial factor that has the effect of preventing bacteria growth **(Radford and Board, 1993).**

Table (9): Changes in some microbial groups (cfu/g) of mayonnaise samples during storing at 4°C for 21 days

Treatments	Storing periods (days)	C	T1	T2	T3	T4	LSD
Total bacterial count	1	$11.00\pm1.00^{A,d}$	ND	$6.67\pm2.52^{B,d}$	$4.67\pm0.58^{BC,d}$	$2.33\pm0.58^{C,c}$	2.66
	7	$14.67\pm0.58^{A,c}$	ND	$11.00\pm1.00^{B,c}$	$10.67\pm2.08^{B,c}$	$7.33\pm1.15^{C,b}$	2.49
	14	$21.33\pm1.53^{A,b}$	$4.67\pm0.58^{C,b}$	$19.33\pm3.06^{A,b}$	$17.67\pm3.79^{A,b}$	$9.00\pm1.00^{B,b}$	4.25
	21	$40.00\pm2.00^{A,a}$	$17.33\pm2.08^{D,a}$	$27.33\pm1.15^{B,a}$	$24.00\pm1.00^{BC,a}$	$20.67\pm2.52^{CD,a}$	3.35
		2.61	3.46	4.00	4.21	2.82	
Salmonella spp	1	ND	ND	ND	ND	ND	-
	7	ND	ND	ND	ND	ND	-
	14	ND	ND	ND	ND	ND	-
	21	ND	ND	ND	ND	ND	-
Staphylococcus aureus	1	ND	ND	ND	ND	ND	-
	7	ND	ND	ND	ND	ND	-
	14	ND	ND	ND	ND	ND	-
	21	ND	ND	ND	ND	ND	-
Yeasts & molds	1	ND	ND	ND	ND	ND	-
	7	$19.33\pm1.53^{A,c}$	$1.67\pm0.58^{D,c}$	$7.67\pm1.53^{B,c}$	$4.67\pm0.58^{C,b}$	ND	2.17
	14	$24.00\pm1.00^{A,b}$	$4.67\pm1.15^{D,b}$	$13.00\pm1.00^{B,b}$	$11.67\pm1.15^{B,a}$	$7.33\pm0.58^{C,a}$	1.82
	21	$39.67\pm3.01^{A,a}$	$8.67\pm1.53^{D,a}$	$18.67\pm2.08^{B,a}$	$14.33\pm2.51^{BC,a}$	$10.33\pm3.51^{CD,a}$	4.79
		4.11	2.31	3.20	3.26	5.71	

ND: Not detected

C: Mayonnaise with no antioxidants. T1: Mayonnaise treated by TBHQ with level 200 ppm.

T2, T3, T4: Mayonnaise treated by 0.5, 1.0 and 1.5 % ethanolic chard leaves extract, respectively.

The superscript capital letters are differed significantly ($P \leq 0.05$) in the same row.

The superscript small letters are differed significantly ($P \leq 0.05$) in the same column.

Conclusion

From the obtained results, it could be concluded that ethanolic chard leaves extract had a high phenolic content and showed high antioxidant activity. The addition of chard leaves extract at a level of 1.5% during the processing of mayonnaise improved its oxidative stability. Thus, chard leaves extract can be used instead of artificial antioxidants to retard oxidation and expand the mayonnaise shelf-life.

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تقييم جودة المايونيز المدعم بالمستخلص الإيثانولي لأوراق السلق

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الملخص العربي:

تلعب المستخلصات النباتية دوراً هاما في زيادة فترة الصلاحية للأغذية كاملة الدسم، ويهدف هذا البحث الى دراسة تأثير إضافة المستخلص الإيثانولي لأوراق السلق على الخواص الكيميائية والفيزيائية والحسية والميكروبيولوجية للمايونيز، وقد تم اضافة مستخلص أوراق السلق أثناء تصنيع المايونيز بنسب 0.5 و1.0 و1.5٪ من وزن المايونيز، بعد ذلك تم مقارنة هذه العينات مع عينة مايونيز مدعمة بأحدى مضادات الأكسدة الصناعية (مادة TBHQ) وأخرى بدون أى مضادات أكسدة كعينة كنترول، وأظهرت النتائج أن أوراق السلق المجففة تحتوي على نسب مرتفعة من البروتين والرماد والفينولات الكلية، وقد اعطى مستخلص اوراق السلق نشاطا عاليا كمضاد للأكسدة ٩٠,١١ ± ١,٢١٪ مقارنة بـ ٩٥,٢٢ ± ٠,٨٧٪ لمادة TBHQ. ولم يؤثر إضافة مستخلص أوراق السلق للمايونيزعلى تركيبه الكيميائي. ولكنه أدى إلى تغير لون المايونيز من اللون الأصفر الفاتح االى اللون البيج وفقد جزء من مظهره اللامع، وسجلت عينات المايونيز المحتوية على ١,٥٪ من مستخلص أوراق السلق انخفاض غير معنوي في معظم درجات التحكيم الحسي ، باستثناء اللون، مقارنةً بعينات المايونيز الكنترول والمحتوية على TBHQ، ولم يلاحظ وجود اختلافات معنوية في قيم TBA بين عينات المايونيز المحتوية على TBHQ والمحتوية على ١,٥٪ مستخلص أوراق السلق خلال فترات التخزين المختلفة لمدة ٢١ يوم، كما زادت قيم كلا من TBA والحموضة والعدد الكلى للبكتيريا وعدد الخمائر والفطريات في جميع عينات المايونيز بتقدم فترات التخزين فى حين انخفضت قيم اللزوجة وpH.

توصي الدراسة بضرورة استخدام مستخلص اوراق السلق بنسبة ١,٥٪ اثناء تصنيع المايونيز لما له من تاثيرات مضادة للاكسدة طبيعي واطالة فترة الصلاحية وذلك بدلا من مضادات الاكسدة الكميائية.

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