Middle East Journal of Applied Sciences
ISSN 2077-4613

Volume : 05 | Issue : 04 | Oct.-Dec. | 2015
Pages: 1082-1090

Quality Characteristics of Rice Biscuits Sweetened with Carob Powder

Ola S. Ibrahim, Amera T. Mohammed and Hayat H. Abd-Elsattar

Food Tech. Res. Inst. Agric. Res. Cent., Giza, Egypt.

ABSTRACT

The current study aims to produce biscuit from rice flour sweetened with carob powder (*Ceratonia Siliqua)*, which has better nutritional and healthful functional value. Rice biscuit supplemented with 20%, 25%, 30%, 35%, and 40 % of carob powder. Chemical composition, minerals (Mn, Zn, Fe, Ca, Na, Mg and K) and total phenol content were determined. Physical and sensory characteristics were evaluated. The results showed that addition of carob powder to rice biscuit at 35% ratio resulted in the highest overall acceptability and characterized with high content of crude fiber, protein, Na, K, Ca and Fe; however, lower value of carbohydrates content compared with control. At the end of storage period,(six-months) the peroxide value showed that minimum value for rice biscuit supplemented with 25% followed by 30% and 20% of carob powder. The microbiological quality showed that an increase in total count, yeast and mould in rice biscuit supplemented with 35% and40 % and a decrease in those of 25 % and 30 % of carob powder compared with the control due to the differentaddition amount of carob powder. Therefore, it could be recommended to utilize the carob powder as a natural sweetened also, to enhance the nutritional value.

***Keywords**:* Carob powder, rice biscuit, chemical composition, Minerals, Physical properties, Sensory characteristics, antioxidant, and anti microbial.

Introduction

The carob *(Ceratonia Siliqua*) is a perennial leguminous tree, native to the Mediterranean basin and southwest Asia, it belongs to family febaceae and caesalpinioideae sub-family (Naghmouchi *et al*., 2009). It had been cultivated throughout the Mediterranean region for approximately 4000 year (Santos*et al.*, 2005). Total area is approximately 200,000 hectares and the yield depends on cultivar, region and cultural practices (Makris and Kefalas, 2004). Carob is typically dried or roasted, and is mildly sweet. In powdered, chip, or syrup from, it is used as an ingredient in cakes and cookies, and is used as a substitute for chocolate. Crushed pods may be used to make a beverage; compote, liqueur, and syrup are made from carob in Turkey, Malta, Portugal, Spain and Sicily (Burg, 2007). Carob powder is a natural sweetener with flour and appearance similar to chocolate; therefore, it is often used as cocoa substitute. The advantage of using carob as a chocolate resides in that carob is an ingredient free from caffeine and theobromine (Bengoechea *et al.*, 2008). Seeds endosperm contains polysaccharides (galactomannan) which called carob gum (Batlle and Tous,1997). Locust bean gumis commonly used in the food industry as dietary fiber, thickening and foaming agent, emulsifier, and stabilizer as a drug delivery (Mirhosseini and Amid, 2012).

Carob pod mainly consists of pulp (90 %), seeds (10 %), which is rich in sugars (48–56%), but it also contains a large amount of condensed tannins (16–20 %) (Batlle and Tous, 1997). Avallone *et al*. (1997) reported that carob pod characterized by a high content of carbohydrates (45%, with sucrose more than 30%), appreciable amounts of protein (3%), low levels of fat (0.6%) and also a high tannin content is present in carob pod composition, which limits the consumption by cattle because of reduced digestibility (Priolo *et al*., 2000). Khlifa *et al*.(2013) reported that the main values of chemical composition of carob kibbles is calculated on six essays were as follow (g/100g dry matter) protein (2.74±0.03), ash (3.0± 0.03), total and reducing sugar(83.7, 512.5),sucrose (44.64 ±0.49), fructose (7.2 ± 0,2), glucose (2.2 ±0.2), crude fiber (6.9± 0.06), tannins (6.7 ± 0.2) and total poly phenol (17.0 ±0.5); and minerals (mg/100g), P: 76.22, K: 1003.5, Ca: 268.57, Mg: 96.43, Fe:2.1 and Zn: 0.45. Kumazawa *et al*. (2002) reported that carob pod crude poly phenol had high antioxidant activity comparable to that of authentic poly phenol compounds. Especially, it is apparent that carob pod crude poly phenol has strong effect against the discoloration of β-carotene. Similarly, Papagiannopoulos *et al. (*2004) showed that carob fiber and carob flours have high anti oxidative activity expressed with a high DPPH radical scavenging activity. The antioxidant activity of the carob pod is attributed to the presence of phenolic compounds (Owen *et al*, 2003 and, Faik *et al,* 2007). In addition, antioxidants have been widely used in the food industry to prolong shelf life. However, there is a widespread agreement that some synthetic antioxidants such as butyl hydroxyl anisole and butyl hydroxyl toluene (BHA and BHT respectively) need to be replaced with natural antioxidants because of their potential health risks and toxicity. Therefore, the search for antioxidants from

Corresponding Author: Ola S. Ibrahim, Food Tech. Res. Inst. Agric. Res. Cent., Giza, Egypt.
E-mail: dr.ola-salah-farid@hotmail.com - amirataha99@yahoo.com

natural sources has received much attention, and efforts have been made to identify new natural resources for active antioxidant compounds. In addition, these naturally occurring antioxidants can be formulated to give nutraceutical, which can help to prevent oxidative damage from occurring in the body (Kahl and Kappus,1993). Fruits and leaves of carob tree contain phenolic compound and exhibit, antioxidant, antiproliferative and anti microbial activity. (Custódio *et al*., 2011). Some medicinal plants emerge as alternative source for new that natural antimicrobial agent (Abdallah, 2011), it is known that phytochemical compounds which have curative properties (Mallikharjuna *et al*., 2007). The major phytochemical detected in *Ceratonia Siliqua* L. are polyphenols including condensed and hydrolysable tannins, phenolic and flavonoids and flavonoidal glycosids suggesting potential antibacterial and cytotoxic activities (Custódio *et al*., 2011). Methanolic extracts of *Ceratonia Siliqua* L have antioxidant and antimicrobial properties (Bijen and Tuba, 2002) and it showed a strong action for Entreococcus, Escherichia coli and Staphylococcus arueus (Ben-Hsouna *et al*., 2011).

Rice competes closely with wheat as the world most important food crop. It isgrown in more than 100 countries including Egypt, when compared with other cereals; it has the highest food yield. Regarding to wheat and maize, rice supplies more than half of all the calories for human consumption.White rice flour is usedin many gluten-free formulas. It is available in several different textures, from regular to fine grinds. Chemical composition of milled rice flour for rice cultivars ranged from 6.6 to 9.3 %, 0.18 to 0.51%, 9.0 to 11.0 % and 19.6 to 27.0% for crude protein, Crude fat, moisture and amylose, respectively (Cameron and Wang, 2005). White rice contained 173.0, 28.7 and 24.5 mg / Kg for Ca, Fe and Zn, respectively (Liang *et al*., 2009). Broken rice flour had 7.68% protein, 0.7% fat, 0.27% crude fiber, 0.36% ash, and 90.81% carbohydrate (Dalia, 2006).

Celiac disease (CD) is an autoimmune and chronic disorder in which the mucous membrane of the small intestine is damaged in gluten-intolerant individuals. CD is caused by not only a reaction to gliadin in wheat prolamin but also high molecular glutenin and subunits of gluten protein consequents in damage and inflammation to the small intestine and causes malnutrition (Demirkesen *et al.*, 2013). This chronic disease is recognized as long-life disease and the only solution is adherence stickiness to gluten-free products. However, this is not easy as many foodstuffs contain gluten (Motrena *et al*. 2011). In genetically susceptible individuals the ingestion of gluten causes an inappropriate small intestinal immune response characterized by villous atrophy, resulting in malabsorption of proteins, fat, carbohydrates, soluble vitamins, float and minerals especially iron and calcium. Recent studies in Europe, India, South America, Australia and USA Indian estimated mean prevalence of 0.33- 1.06% in children and 0.18-1.2% in adults.(Ballabio *et al*., 2011) .

Gluten-free biscuits are typically round cakes with baking powder, baking soda. It may also refer to cookies or crackers. Biscuits may be regarded as a form of confectionery dried to very low moisture content. Also, biscuit is defined as a small thin crisp cake made from unleavened dough. Biscuits may be classified either by the degree of enrichment and processing or by the method adopted in shaping them. Based on the enrichment criterion, biscuits may be produced from hand dough, soft dough or from batter. (Agu *et al*., 2007).

The aim of this study was to produce biscuits from rice flour sweetened with carob powder (*Ceratonia Siliqua)* which more suitable for Celiac disease and study it's properties: physical, chemical, microbial quality, peroxide value and sensory properties of the developed products.

Materials and Methods

Materials*:*

Carob pods (*Ceratonia Siliqua*) were obtained from local market and ground to particles around 0.45 mm after removed seeds, then kept in polyethylene in the refrigerator till using. Rice flour, Sugar powder, shortening, eggs, vanillin and baking powder were purchased from local market, Giza, Egypt. Total plat count agar media was obtained from Canada pronodisa, Spain. Maccounkey agar media was obtained from Bilolilife, Milano Italy. Yeast and mould agar media were obtained from Difco^{TM} Co. (U.S.A).

Methods:

Production of biscuit:

Biscuit Formula and Ingredients:

Biscuit dough was prepared according to the formula presented in Table (1), which described by (Sayed, 2011). Rice biscuit supplemented with carob powder were prepared using the same formula except for replacing the rice flour with carob powder at different level from 20% to 40%.

Dough preparation:

Powdered sugar and shortening were creamed in Braun Mixer with a flat beater for 2 minutes at high speed. Egg and vanillin were added to the cream and mixed for 5 minutes to obtain a homogenous cream. Baking powder was added slowly to the flour and mixed for 2 minutes at low speed to obtain biscuit dough. Then, it was sheeted to a thickness of about 3mm, the sheeted dough was cut into round shape using a 45 mm diameter. The biscuits were baked at 170 C° for 12 min. Biscuits were kept at room temperature for 75 minutes after baking, then packed in polyethylene packet and stored at room temperature (± 25C°) for 6 months (Sayed, 2011 and Manohar and Rao,1997).

Table 1: Formula of rice biscuit supplemented with carob powder (g).

Treatment / Ingredients	Rice flour	Carob powder	Sugar powder	shortening	Baking powder	eggs	vanillin
CO	100	-	30	30	3	15	1
RFCP 20%	80	20	19.2	30	3	15	1
RFCP 25%	75	25	16.5	30	3	15	1
RFCP 30%	70	30	13.0	30	3	15	1
RFCP 35%	65	35	11.0	30	3	15	1
RFCP 40%	60	40	8.3	30	3	15	1

CO = control (100 rice flour), RFCP 20% = Rice flour supplemented with 20% carob powder, RFCP 25% = Rice flour supplemented with 25% carob powder, RFCP 30% = Rice flour supplemented with 30% carob powder, RFCP 35% = Rice flour supplemented with 35% carob powder, RFCP 40% =Rice flour supplemented with 40% carob powder, Total soluble sugar in carob powder 54.2 % .

Analytical methods:

Crude protein, crude fat, ash, crude fiber and moisture contents were analyzed according to the procedures described in (AOAC,2005). Carbohydrates were calculated by difference. Minerals content were determined according to (AOAC, 2005) using atomic absorption spectrophotometer model 3300 (USA).Total soluble sugar were determined according to the method described by (Miller, 1959).Total phenols were determined in sample by the Folin-Ciocalteau using the method outlined by (Kaluza *et al*., 1980).

Physical Evaluation of Biscuits:

Physical characteristics were determined according to the method described by (Manohar and Rao, 1997) Biscuits were evaluated for thickness (cm), width (cm), spread ratio and spread factor. Five biscuits were used for the evaluations and averages were recorded. The spread ratio and spread factor were calculated according to the following equations:

Spread ratio= width (diameter) / height (thickness)

Spread factor = Spread ratio of sample / Spread ratio of control ×100

The average weight of five biscuits was recorded. Volume (cm^3) was determined by displacement of rapeseed.The density was calculated and expressed as g /cm^3.

Sensory Evaluation of Biscuits:

Sensory characteristics of biscuits were evaluated at zero time and after 6 month according to the method described by (Manohar and Rao, 1997) which carried out by panel of ten experienced guides from the staff of the food Tech. Res. Institute, Agric., Res. Center, Giza, Egypt. Assigning scores for various qualities attributes such as: color (20), taste (20), texture (20), crust appearance (20), odor (20), and overall acceptability (100).

Antioxidant activity:

Antioxidant activity was determined by free radical scavenging activity according to the method described by (Miliauskas, *et al*, 2004).

Peroxide value:

Peroxide value was determined according to the method out lined in(AOAC, 2005)total bacteria count, yeast, mould and coliform group were determined according to the method described in(APHA, 1992)

Statistical Analysis:

The obtained data were exposed to analysis of variance (ANOVA). Duncan's multiple range tests at ($P \leq 0.05$) level was used to compare between means.

Results and Discussion

Chemical Composition of carob powder and rice flour:

Chemical composition and minerals content of carob powder and rice flour were presented in Table (2). The carob powder sample contained low level of fat (1.07%). The crude fiber and protein contents were 6.82 % and 6.89 %, respectively, the carbohydrate content was 82.56 % which was extremely high. The data are in good agreement with (Owen *et al,* 2003) who stated that the protein content of carob was 8.0%. Eman and Awlya (2012) found that protein was recorded a high amount (9.32 %), also the carbohydrate content represented 57.06% and the fat 5.29% ; Kamal *et al* (2013) and Khlifa *et al* (2013) found that the carob powder contained low level of fat 0.99- 1.99%, protein 2.68-6.34%, crude fiber 6.79-7.30% and carbohydrate 75.92% .

The data revealed that the carob powder is considered as a rich source of Fe, Ca, Na, K and Mg, while Mn and Zn were found in lowest values (14.31 and 7.84 mg/kg) compared with other minerals. The data are in good agreement with (Faik *et al*, 2007) who reported that the major minerals were K, Ca, P and Mg (970, 300, 71 and 60 mg/100g, respectively) and traces were Zn, Mn and Cu (0.75, 1.29 and 0.85 mg/100g, respectively). Eman and Awlya, (2012) showed that K, Ca and Na were the highest minerals in carob powder (1486.25, 220.41 and 110.93 mg/100g, respectively), while Fe and Zn were the lowest mineral (2.23 and 0.62 mg/100g, respectively). Kamal *et al* (2013) found that the carob powder is considered as a rich source of Fe, Ca, Na, K and P(381.80, 2123.0, 505.97, 8637.64 and 2255.21 mg/kg, respectively), while the trace element Cu and Mn (4.84 and10.24 mg/kg ,respectively) .

Table 2: Chemical Composition (%) and Minerals Content (mg / kg) of Carob powder and Rice flour .

Analysis	Carob powder	Rice flour
Moisture	5.91	9.82
Ash	2.65	0.80
Protein	6.89	7.95
Crud fiber	6.82	0.42
Crud fat	1.07	0.30
Total Carbohydrate	82.56	90.53
Total soluble sugar	54.2	-
Na	512.91	3.85
K	11315.4	621.5
Mg	1634.34	510.2
Ca	2285.76	90.5
Fe	147	2.2
Mn	14.31	1.25
Zn	7.84	2.80

The same table showed that the rice flour had the highest value of total carbohydrate content (90.53%), low level fat and crude fiber (0.30 and 0.42%, respectively) and protein content was 7.95 % .These result are in accordance with those reported by(Cameron and Wang, 2005) who found that milled rice flour had (6.6-9.3%) protein and (0.18-0.5%) fat . Dalia (2006) reported that broken rice flour contained 7.68% protein, 0.7% fat, 0.27% crude fiber, 0.36% ash and 90.81% carbohydrate.

Concerning to the minerals content of rice flour, it could be reported that Fe, Ca, Zn, Mg, Na and K content was 2.2, 90.5, 2.80, 510.2, 3.85 and 621.5 mg/kg, respectively. These results are in agreement with those reported by Dalia (2006) who found that the broken rice flour had Fe 1.3 mg/100g, Mg 30 mg/100g, Zn 0.38 mg/100g, Ca 13.2 mg/100g, Mn 0.24 mg/100g, Na 1.4 mg/100g and K 51.86 mg/100g.

The Total phenol and anti-oxidant activity of carob pod powder and rice flour:

The data of total phenol and anti-oxidant activity of carob powder and rice flour are presented in Table (3). The carob powder contained 1256.6 mg/100g of total phenol. Another study of carob showed that contained 13.51 mg/g Gallic acid equivalent (GAE/g) of total phenolics (Faik *et al,* 2007). Stephanie *et al*. (2009) found that total phenol was 23.58 mg/g Gallic acid equivalent (GAE/g), and El Mostapha *et al (*2010)showed that the phenolic content in carob grown in different regions of morocco were ranged from (9.15 to 55.73 mg/g gallic acid). On other hand, the anti-oxidant activity of carob powder in this study was 84.9% of DPPH, the data are in good agreement with (Hichem *et al,* 2013) who reported that the antioxidant power for pulp in water and methanol extract of carob was 40.03 and 23.26 % respectively, compared to 79.41% of inhibition for trolox, a well-known antioxidant molecule. The antioxidant capacity of carob extracts is mainly related to their higher level of phenolic compound (Rodrigoand Bosco, 2006; and Seifried *et al*, 2007). Stephanie *et al* (2009) showed that the antioxidant capacity of carob pods was 7.70% of DPPH inhibition. Antioxidant properties of phenolic compounds are directly linked to their structure. Indeed, phenolics are composed of one or more aromatic rings bearing one or more hydroxyl groups and are therefore potentially able to quench free radicals by forming resonance-stabilized phenoxyl radicals (Rice-Evans *et al*, 1996; and Bors and Michel, 2002) .

Table 3: Total phenol and anti-oxidant activity of carob powder and rice flour.

Analysis	Carob powder	Rice flour
Total phenol (mg/100g)	1256.6	115
Anti-oxidant activity %	84.9	ND

ND = not detected

The same table showed that the rice flour had 115mg /100 g total phenol. These results are in agreement with those reported by Shen *et al* (2009) found that the total phenolic content in white rice was 151.8 mg/100g.

Sensory Characteristics of Biscuits:

Sensory characteristics of the studied rice biscuit as influenced by the incorporation of 20, 25, 30, 35, and 40% carob powder are outlined in Table (4). The data revealed that supplementation of rice biscuit with carob powder improved some sensory characters compared with control; supplementation with 35% carob powder recorded the highest score of all studied characters. Therefore, it could be recommended at 35% carob powder supplementation produced biscuits with good quality and acceptable sensory quality attributes. Kamal *et al* (2013) found that 10% and 20% fortified biscuits with carob powder improved all studied sensory characteristics. However, the best scores of all studied were recorded for 20%-fortified biscuits with carob powder. Sensory evaluation continued during the storage period with showed a slight variation (non-significant) and when the peroxide value reached to the recommended value for the deterioration (10.0 meqi/kg fat), sensory evaluation stopped for the chemically deterioration.

Table 4: Sensory characteristics of carob rice biscuits.

Treatments	Crust appearance (20)	Color (20)	Taste (20)	Odor (20)	Texture (20)	Overall Acceptability (100)
CO	17.60^{b} ±1.42	18.05^{b} ±1.06	17.15^{ab} ±2.86	17.60^{a} ±2.75	18.20^{a} ±1.31	88.60^{ab} ±5.87
RBCP 20%	17.30^{b} ±1.15	18.0^{b} ±0.66	17.40^{ab} ±1.62	17.85^{a} ±2.96	18.05^{a} ±1.67	88.60^{ab} ±4.23
RBCP 25%	17.50^{b} ±0.97	18.30^{b} ±0.78	16.60^{ab} ±1.32	17.45^{a} ±2.71	17.30^{a} ±1.76	87.15^{b} ±4.56
RBCP 30%	17.85^{b} ±1.05	18.20^{b} ±0.91	16.25^{b} ±2.46	17.85^{a} ±2.92	17.30^{a} ±1.94	87.45^{b} ±5.73
RBCP 35%	18.90^{a} ±0.99	19.10^{a} ±0.73	18.30^{a} ±0.78	18.30^{a} ±1.76	18.20^{a} ±1.85	92.80^{a} ±4.37
RBCP 40%	17.85^{b} ±0.88	18.25^{b} ±0.88	17.90^{ab} ±1.54	18.10^{a} ±1.95	17.45^{a} ±2.03	89.55^{ab} ±5.79
L.S.D	0.98	0.75	1.70	2.25	1.59	4.61

CO = control (ricebiscuit),RBCP 20% = Rice biscuit supplemented with 20% carob powder, RBCP 25% = Rice biscuit supplemented with 25% carob powder, RBCP 30% = Rice biscuit supplemented with 30% carob powder, RBCP 35% = Rice biscuit supplemented with 35% carob powder, RBCP 40% =Rice biscuit supplemented with 40% carob powder

Physical Characteristics of Biscuits:

Physical characteristics of rice biscuits and fortified rice biscuits with carob powder such as diameter, thickness, density, spread ratio and spread factor are presented in Table (5). The data recorded a gradual increment of spread ratio of fortified rice biscuits with carob powder. Considering the spread factor of control biscuits as 100, but it increased in fortified rice biscuits with carob powder. The thickness of the biscuits with carob powder decreased than control biscuits.

Table 5: Physical characteristics of rice biscuits and carob rice biscuits.

Treatments	Diameter (mm)	Thickness (mm)	Density g/cm³	Spread ratio D/T	Spread factor %
CO	42.31	6.50	1.11	6.51	100
RBCP 20%	42.95	6.49	0.83	6.62	101.68
RBCP 25%	42.50	6.35	0.81	6.69	102.76
RBCP 30%	42.25	6.34	0.76	6.66	102.30
RBCP 35%	42.04	6.35	0.70	6.62	101.68
RBCP 40%	41.28	6.18	0.69	6.67	102.45

CO = control (Rice biscuit), RBCP 20% = Rice biscuit supplemented with 20% carob powder, RBCP 25% = Rice biscuit supplemented with 25% carob powder, RBCP 30% = Rice biscuit supplemented with 30% carob powder, RBCP 35% = Rice biscuit supplemented with 35% carob powder, RBCP 40% =Rice biscuit supplemented with 40% carob powder, Thickness of 5 biscuits-diameter of 5 biscuits
Spread ratio = Width / Thickness, Spread factor = Spread ratio of sample /Spread ratio of control×100.

Chemical Composition of Biscuits:

Chemical composition and minerals content of rice biscuits supplemented with carob powder are given in Table (6).

The data revealed that incorporation of 20, 25, 30, 35 and 40% of carob powder in rice biscuits increased in crude fat, ash, crude fiber and all studied minerals (Na, K, Fe and Ca) except Mg, while decreased in protein and carbohydrate. Kamal *et al* (2013) reported that 10% and 20 % carob powder fortified biscuits increased crude fat, ash, crude fiber, Ca and K.

Phenolic compounds of rice biscuits and fortified rice biscuits with carob powder (35%) are given in Table (7). The data revealed that the phenols and Anti oxidant activity increased in rice biscuits fortified with 35% of carob powder. Kamal *et al* (2013) found that 10 and 20% carob powder fortified biscuits recorded the highest phenolic compound.

Table 6: Chemical Composition (%) and Mineral Contents (mg/kg).

Chemical analysis%	Control	RBCP 20%	RBCP 25 %	RBCP 30%	RBCP 35 %	RBCP 40 %
Moisture	5.22	5.51	5.39	5.60	5.41	5.65
Ash	1.35	2.11	2.40	2.21	2.33	2.38
Protein	8.10	6.52	6.45	6.62	6.65	6.74
Crude fiber	0.95	1.11	1.35	1.41	1.52	1.58
Crude fat	27.50	27.41	27.65	28.33	28.57	28.77
Total Carbohydrate	62.10	62.85	62.15	61.43	60.93	60.53
Na	3.23	204.69	168.78	168.27	205.71	165.45
K	582	1150.19	1183.1	1391.95	1538.25	1544.05
Mg	400	396.39	438.95	338.29	361.99	343.87
Mn	1.07	2.45	2.28	2.59	2.28	2.18
Zn	3.75	9.17	8.71	7.99	8.45	7.40
Ca	107.3	1797.72	1809	1820	1853	1910
Fe	2.31	9.65	10.86	15.81	25.14	26.38

CO = control (Rice biscuit), RBCP 20% = Rice biscuit supplemented with 20% carob powder, RBCP 25% = Rice biscuit supplemented with 25% carob powder, RBCP 30% = Rice biscuit supplemented with 30% carob powder. RBCP 35% = Rice biscuit supplemented with 35% carob powder, RBCP 40% =Rice biscuit supplemented with 40% carob powder.

Table 7: Total phenol andanti-oxidant activity of biscuits.

Treatments	Total phenol	Antioxidant activity
Control	ND	ND
RBCP 35%	659.71	0.78

Control: rice biscuits, RBCP 35% : biscuit of 35 % carob powder supplemented rice biscuits .
ND = non detected

The effects of Carob powder on the peroxide value (pv) of the different formulas during storage period (6 months) at room temperature 25±2˚C:

Table (8): showed the peroxide value of the extracted oil from the different formulas compared with the control during storage period (zero up to 6 month) at room temperature (25± 2˚C) . From the results, the peroxide value ranged from 0.11 to 0.16 mequiv./kg fat at zero time for all formulas .An increase was found after three month. The deterioration of the formulas due to the increasing (pv) was found after 4 months for the control which recorded 10.9 mequiv./kg fat and 6 months for each rice biscuit supplemented with 35% and 40% of carob powder which recorded 13.0 and 20.0 mequiv./kg fat, respectively. At the end of storage period (6 months), the peroxide value showed itś minimum value for rice biscuit supplemented with 25% followed by 30% and 20% of carob powder which recorded 7.0, 8.02, and 9.25 mequiv./ kg fat, respectively after 6 months of storage period. It is worth mentioning that deterioration was calculated when the (pv) reached 10 mequiv./kg fat according to food chemical codex (2003). From the previous results; the increasing of the peroxide value in some rice biscuit supplemented with carob powder may be due to the auto oxidation or others ,such as water activity (Aw) (Baker, 2002) .

Table 8: The effect of carob powder on peroxide value of rice bicuits and carob rice biscuits during storage period at room temperature ±25Cº up to 6 months.

Treatments		Storage period (month)					
	Zero time	1	2	3	4	5	6
CO	0.160	0.570	2.136	7.30	10.9	-	-
RBCP 20%	0.150	0.390	1.480	2.90	5.2	6.40	9.25
RBCP 25 %	0.132	0.220	0.410	1.19	2.9	3.40	7.00
RBCP 30%	0.114	0.192	0.318	2.00	4.5	5.75	8.02
RBCP 35%	0.114	0.210	0.355	2.80	5.9	6.65	13.00
RBCP40%	0.110	0.130	0.219	3.20	6.0	8.20	20.00

*CO = control (rice biscuits), RBCP 20% = Rice biscuit supplemented with 20% carob powder, RBCP 25% = Rice biscuit supplemented with 25% carob powder, RBCP 30% = Rice biscuit supplemented with 30% carob powder, RBCP 35% = Rice biscuit supplemented with 35% carob powder, RBCP 40% =Rice biscuit supplemented with 40% carob powder, * The peroxide value was calculated as mequiv.of O2/ kg oil*

Also, the anti oxidant properties of carob powder *(Ceratonia Siliqua)*, which was added by different amounts may be caused a decrease in the peroxide value in other formulas. These results agreement with (Rodrigo and Bosco , 2006; and Seifried *et al*, 2007) who reported that the antioxidant capacity of the carob extract is mainly related to their higher level of phenolic compound in this fraction . These latter are well known for their ability of scavenging free radicals such as superoxide radical (O_2), hydroxyl radical (OH) and others(ROS). Also a positive correlation between phenolic compounds and antioxidant capacity is common in the majority of natural extracts(Chon *et al*., 2009 and Hamed *et al*., 2010) . More recent studies indicated that carob pods contain1.9 mg/g of total polyphenols, 0.28 mg/g of proanthocyanidins and 0.1 mg/kg of hydrolysable tannins(gallo- and ellagitannins). Another examination of carobs showed their contents in total phenols and total flavanols to be

19.2 and 4.37 g per 100 g, respectively (Kumazawa *et al*., 2002). Furthermore, carob pods were reported to contain 6.1 % of total polyphenol, and chemical degradation of tannins produced flavanols including catechin, epicatechin, epigallocatechin, epigallocatechin gallate, epicatechin gallate, along with simpler phenolics such as phloroglucinol pyrogallol, catechol, and gallic acid (Makris and Kefalas, 2004).

The effect of carob powder on the microbial quality for all formulas during storage period (6 months) at room temperature (25±2 ˚C):

Table (9) showed the resultant total bacteria count, *E. coli* , and Yeast and mould in different formulas (form 1 to 6) of rice biscuits supplemented with C.P during storage period (6 months) at room temperature (25±2 Ć). The results fixed at the second dilution for total bacteria count and yeast and mould and at the first dilution for coliform group. The microbial quality showed the presence of total count at zero time 0.1, 0.1, ND,ND, 0.2 and 0.2 for the formulas under study. Yeast and mould not detected in all studied formulas except (for.5 and for.6) which recorded 0.1cuf for each one. Moreover, *E. coli* not detected in all formulas during storage period.

Table 9: The effect of carob powder on the microbial quality of Rice biscuit and carob ricebiscuit during storage period at room temperature ±25Cº up to 6 months.

Storage period (Month) Treatment	Zero time			6 months		
	T.C**	Y&M**	*E. coli**	T.C**	Y&M**	*E. coli**
Control	0.10	ND	ND	40.0	10	ND
RBCP 20%	0.10	ND	ND	30.0	0.7	ND
RBCP 25 %	0.0	ND	ND	1.0	ND	ND
RBCP 30%	0.0	ND	ND	7.0	0.3	ND
RBCP 35%	0.20	0.10	ND	50.0	19.0	ND
RBCP 40%	0.20	0.10	ND	80.0	70.0	ND

CO = control (rice biscuits),RBCP 20% : biscuit of 20% carob powder supplemented rice biscuits, RBCP 25% : biscuit of 25% carob powder supplemented rice biscuits, RBCP 30%: biscuit of 30 % carob powder supplemented rice biscuits, RBCP 35% : biscuit of 35 % carob powder supplemented rice biscuits, RBCP 40% : biscuit of 40 % carob powder supplemented rice biscuits, T.C :Total bacteria count at the first dilation , Y&M : Yeast and mould at (10^{-2}) dilation, C.G : Coliform group at (10^{-2}) dilation.
ND : Not detected

After 6 months of storage, total count showed itś maximum value (80 cfu) for rice biscuit supplemented with 40 % followed by 35 % , 20 %, 30 % and 25 % of carob powder which recorded 50, 30, 7.0 and 1.0 cuf , respectively compared with the control (40 cuf). Yeast and mould showed itś maximum value in rice biscuit supplemented with 40 % of carob powder (70 cfu), which was about 7.0, 100, 233.3 and 3.68 times as that of control followed by 20 % , 30 % and 35 % of carob powder , respectively. While rice biscuit supplemented with 25 % of carob powder showed no detectable yeast and mould. The pervious results revealed an increase in total count and yeast and mould in rice biscuit supplemented with 35 % and 40 % and a decrease in those of 30 % and 25 % of carob powder compared with the control related to the different addition amounts of carob powder *(Ceratonia Silque*) which inhibit the growth of the microbes due to the presence of some phytochemicals (polyphenols, flavonids).

These results agreement with (Bijen and Tuba 2002. and Anis *et al*., 2011) who reported that methanol extract of (*Ceratonia Silque*) showed a strong action on *Enterococcus, Escherichia coli* and *Staphyloc occus Aureus*. The strong antimicrobial activity of (*Ceratonia Silque*) against the microorganisms could be attributed to the presence of high percentage of hydrocarbon (51.06%), monoterpene (0.9%), and oxygenated monoterpene (1.19%) (Sivropoulou *et al*., 1997 and Yangui *et al*., 2009).

Conclusion:

In conclusion, carob pod powder may actually be regarded as functional food, cheap source of natural polyphenolic. Meanwhile, it also supplies people with a good healthy dose of fiber. The soluble fibres in particular are thought to exert a preventative role against heart disease, as they appear to have the ability to lower serum cholesterol. The polyphenols have antioxidant activity, which mainly enhances the prevention or delay the oxidative damage caused by free radicals. Consequently, polyphenols are involved in protection against several diseases (cardiovascular and neuronal),. As a result, carob powder should be increasing interest as an ingredient in the food industry such as functional and healthy foods formulations as Biscuits, bread, and cakes for some patients.

Reference

AOAC, 2005. Official Methods of AOAC 18th Ed., 2005 current through revision I, Basic sensory methods for food Evaluation. IDRC, Ottawa Ontario, Canada.

Abdallah, E.M., 2011. Plants: An alternative source for antimicrobials. J Appl Pharma Sci., 1: 16-20.

Agu, H.O., J.A. Ayo, A.M. Paul and F. Folorunsho, 2007. Quality characteristics of biscuits made from wheat and African bread fruit (Treculia Africana). Nigerian food Journal, Vol.25, No.2.

APHA, 1992. American Publ. Health Assoc, Standard methods for the examination of dairy product.APHAINC.16 the Ed, Washington D.C.

Anis, B. H., T. Mohamed, M. Riadh Ben, M.J. Raoudha, D. Mohamed and Samir, 2011. Chemical composition, cytotoxicity effect and antimicrobial Activity of *Ceratonia siliqua* essential oil with effects against *Listeria* inoculated in preservative minced beef meat. International Journal of Food Microbiology, 148: 66-72.

Avallone, R., M. Plessi, M. Baraldi and A. Monzani, 1997. Determination of chemical composition of carob (*Ceratonia siliqua*): protein, fat, carbohydrates and tannins. J. Food Compos. Anal., 10: 166-172.

Baker, G.L., 2002. Flavor formation and sensory perception of selected peanut genotype (*Arashis hypogea* L) as affected by storage water activity, roasting and planting date. PhD. Diss. Gainesville, Fla. Univ. Florida.

Ballabio, C., F. Uberti, E. Manferdelli, G. Vacca, R. Boggini, C. Catassi and E. Lionetti, 2011. Molecular characterization of 36 oat varieties and invitro assessment of their suitability for diet. Journal of Cereal Science, 54: 110-115.

Batlle, I. and J. Tous, 1997. Carob Tree (*Ceratonia siliqua* L.), Promoting the Conservation and Use of Underutilized and Neglected Crops. 17, Institute of Plant Genetics and Crop Plant Research, Gatersleben International Plant Genetics Resources Institute, Rome.

Ben-Hsouna, A., M. Trigui, R. Ben Mansour, R.M. Jarraya, M. Damak and S. Jaoua, 2011. Chemical composition, cytotoxicity effect and antimicrobial activity of *Ceratonia siliqua* essential oil with preservative effects against Listeria inoculated in minced beef meat. Int J Food Microbiol, 148: 66-72.

Bengoechea, C., A. Romero, A.Villanueva, G. Moreno, M. Alaiz, F. Millan, A. Guerrero and M.C. Puppo, 2008. Composition and structure of carob (*Ceratonia Siliqua* L.) germ proteins. Food chem, 107: 675-683.

Bijen, K. and M. Tuba, 2002. Antimicrobial and Cytotoxic Activities of *Ceratonia siliqua* L. Extracts.Turk J. Biol., 26 :197-200

Bors, W. and C. Michel, 2002. Chemistry of the antioxidant effect of polyphenols. *Ann. N.Y. Acad. Sci.*, *957:* 57–69.

Burg, B., 2007. Good treats for dogs cookbook for dogs: 50 Home-cooked treats for specia occasions. Quarry Books, p.28.

Cameron, D.K. and Y.J. Wang, 2005. A better understanding of factors that affect the hardness and stickiness of long-grain rice. Cereal Chem., 82(2): 113-119.

Chon, S.U., B.U. Heo, Y.S. Park, D.K. Kim and S. Gorinstein, 2009. Total phenolics level, antioxidant activities and cytotoxicity of young sprouts of some traditional Korean salad plants. Plant Foods Hum. Nutr., 64: 25-31.

Custódio, L., A.L. Escapa, E. Fernandes, A. Fajardo, R. Aligué, F. Albericio, N. Neng and J. Manuel, 2011. Phytochemical Profile, Antioxidant and Cytotoxic Activities of the Carob Tree(*Ceratonia siliqua* L.) Germ Flour Extracts. Plant Foods Hum Nutr, 66: 78-84.

Dalia , B.O.D., 2006. Utilization of different cereal flour mixes in the preparation of some baking products. M. Sc. Thesis , Minufiya Univ , Faculty of Agric., 160 pp.

Demirkesen, I., G. Sumnuand, S. Sahin, 2013. Quality of Gluten-Free Bread Formulations Baked Indifferent Ovens. Food and Bioprocess Technology, 6: 746-753.

El Mostapha, R., H. Chicha, S. Abouricha, M. Alaoui, A. A. Bouli, M. Hansali and R.W. Owen, 2010. Determination of Phenolic Composition of Carob pods Grown in Different Regions of Morocco. Journal of Natural Products, 3:134-140 .

Eman, M., Salem and A. Awlya Ohaad Fahad, 2012. Substituting of cacao by carob pod powder In milk chocolate manufacturing. Australian Journal of Basic and Applied Sciences, 6 (3): 572- 578.

Faik, A.A., T. Hülya, A. Sema, J.C. Pedro, A. Manuel, S. Carlos, G. Jiri and S. Miroslav, 2007. Determination of chemical composition of Anatolian carob pod (*Ceratonia siliqua* L.): sugars, amino and organic acids, minerals and phenolic compounds. J. Food Qual., 30: 1040-1055.

Food chemicals codex, 2003. Institute of medical (U.S.).The national academies press. Washington ,D.C. Page, 123.

Hamad, I., Ö. Erol-Dayi, M. Pekmez, E. Önay-Uçar and N. Arda, 2010. Antioxidant and cytotoxic activities of *Aphanes arvensis* extracts. Plant Foods Hum. Nutr., 65: 44-49.

Hichem, S., A. Souli, L. Chehimi, K. Rtibi, M. Amri, J. El-Benna and M. Sakly, 2013. *In vitro* and *in vivo* antioxidant properties of Tunisian carob (*Ceratonia siliqua* L.) Journal of Medicinal Plants Research, 7(2): 85-90.

Kahl, R. and H. Kappus, 1993. Toxicology of the synthetic antioxidants BHA and BHT in comparison with the natural antioxidant vitamin E. *Z.* Lebensm.- Unters Forsch, *196:* 329–338.

Kaluza, W.Z., R.M. McGrath, T.C. Roberts and H.H. Schroder, 1980. Separation of phenolics Of Sorghum bicolor L. moench grain. J. Agric. and Food Chem., 28(6): 1191-1196.

Kamal, M., E. Youssef, M. Moshera, El-Manfaloty and M. Hend Ali, 2013. Assessment of Proximate Chemical Composition, Nutritional Status, Fatty Acid Composition and Phenolic Compounds of Carob (*Ceratonia Siliqua* L.) Food and Public Health, 3(6): 304-308**.**

Khlifa, M., A. Bahloul and S. Kitane, 2013. Determination of Chemical Composition of Carob Pod (Ceratonia Siliqua L) and its Morphological Study. J. Mater. Environ. Sci., 4(3): 348- 353.

Kumazawa, S., M. Taniguchi, Y. Susuki, M. Shimura, M.S. Kwon and T. Nakayama, 2002. Antioxidant activity of polyphenols in carob pods, J. Agric. Food. Chem*.,* 50: 373-377.

Liang, J., B.Z. Han, M.J.R. Nout and R.J. Hamer, 2009. Effect of soaking and phytase treatment on phytic acid , calcium , iron and zinc in rice fractions. Food Chem., 115: 789-794.

Makris, D.P. and P. Kefalas, 2004. Carob pods (*Ceratonia siliqua* L.) as a sources of polyphenolic antioxidants. Food Technology and Biotechnology, 42(2): 105–108.

Mallikharjuna, P.B., L.N. Rajanna, Y.N. Seetharam and G.K. Sharanabasappa, 2007. Phytochemical Studies of Strychnos potatorum L.f.- A Medicinal Plant. E-J Chem., 4: 510-18.

Manohar, R.S. and P.H. Rao, 1997. Effect of mixing period and additives on the rheological characteristics of dough and quality of biscuits. J. of Cereal Sci., 25: 197– 206.

Miliauskas, G., P.R. Vensknionis and T.A. Beek, 2004. Screening of radical scavenging of some medical and aromatic plant extracts. Food chem., 85: 231-237.

Miller, G.L., 1959. Use of dinitro salicylic acid reagent for determination of reducing sugar. Anal. Chem., 31: 426-428.

Mirhosseini, H. and B.T. Amid, 2012. A review study on chemical composition and molecular structure of newly plant gum exudates and seed gums. Food Res Int., 46: 387-98.

Motrena, S.G., M.J. Carvalho, J. Canada, N.B. Alvarenga, F.C. Lidon and B.P. Elisa, 2011. Characterization of Gluten-Free Bread Prepared from Maize, Rice and Tapioca Flours Using the Hydrocolloid Seaweed Agar-Agar. Recent Research in Science and Technology, 3: 64-68.

Naghmouchi, S., M.L. Khouja, A. Romero; J. Tous and M. Boussaid, 2009. Tunisian carob (*Ceratonia siliqua* L.) populations: Morphological variability of pods and kernel. Scientia Horticulturae, 121: 125-30.

Owen, R.W., R. Haubner, W.E. Hull, B. Spiegelhalder, A. Bartsch and B. Haber, 2003. Isolation and Strucidation of the Major Individual Polyphenols in Carob Fibre. Food and Chemical Toxicology, 41(12): 1727-1738.

Papagiannopoulos, M., H.R. Wollseifen, A. Mellenthin, B. Haber and R. Galensa, 2004. Identification and quantification of polyphenols in carob fruits (*Ceratonia siliqua* L.) and derived products by HPLC-UVESI/ MS, *J. Agric. Food Chem.,* 52: 3784-3791.

Priolo, A., G.C. Waghorn, M. Lanza, L. Biondi and P. Pennisi, 2000. Polyethylene glycol as a means for reducing the impact of condensed tannins in carob pulp: effects on lamb growth, performance and meat quality. J. Anim. Sci., 78: 810-816.

Rice-Evans, C.A., N.J. Miller and G. Paganga, 1996. Structure-antioxidant activity relationships of flavonoids and phenolic acid. Free Radical Biol. Med., *20:* 933–956.

Rodrigo, R. and C. Bosco, 2006. Oxidative stress and protective effects of polyphenols: comparative studies in human and rodent kidney. Comp. Biochem. Physiol. C Toxicol. Pharmacol., 142: 317-327.

Santos, M., A. Rodrigues and J.A. Teixeira, 2005. Production of dextran and fructose from Carob pod extract and cheese whey by leuconostoc mesenteroides. NRRL B512(f). Biochemical Engineering Journal, 25: 1-6.

Sayed, A.M.A., 2011. Biochemical and Technological Studies on Barley Talbina. M.Sc. Thesis. Faculty of Agriculture. Assiut University.

Seifried, H.E., D.E. Anderson, E.I. Fisher and J.A. Milner, 2007. A review of the interaction among dietary antioxidants and reactive oxygen species. J. Nutr. Biochem., 18: 567- 579.

Shen, Y., L. Jin, P. Xiao, Y. Lu and J. Bao, 2009. Total phenolics, flavonoids, antioxidant capacity in rice grain and their relation to grain color, size and weight. J. Cereal Sci., 49: 106-111.

Sivropoulou, A., C. Nikolaou, E. Papanikolaou, S. Kokkini, T. Lanaras and M. Arsenakis, 1997. Antimicrobial, cytotoxic, and antiviral activities of *Salvia fruticosa* essential oil. Journal of Agricultural and Food Chemistry, 45: 3197-3201.

Stephanie D., E. X. Vitrac, P. Couti Ere, M. Woillez and J. michel M´erillon, 2009. Comparative Study of Antioxidant Properties and Total Phenolic Content of 30 Plant Extracts of Industrial Interest Using DPPH, ABTS, FRAP, SOD, and ORAC Assays J. Agric. Food Chem*, 57:* 1768–1774.

Yangui, T., M. Bouaziz, A. Dhouib and S. Sayadi, 2009. Potential use of Tunisian *Pituranthos chloranthus* essential oil as a natural disinfectant. Letters in Applied Microbiology, 48: 112-17.