

Effect of Using Thyme, Carrot and Hibiscus to Some Crackers Products on their Nutritional and the Rapeutical Value

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ABSTRACT

The main objective of the present study was carried to evaluate the possibility of enhancing antioxidant activities by using 10% of thyme, carrot and hibiscus powder fortified to crackers as natural colored antioxidants to produce healthy crackers, chemical composition and rheological properties were carried out, determination of phenolic compounds and antioxidant activity% noticed that thyme powder had the highest level of total phenols (12366 mg/100g), followed by carrot powder which was (5087.4 mg/100gm) and hibiscus had (4262.9 mg/100g), while antioxidant activity were 96.7% for thyme, 89.59% for carrot and 86.17% for hibiscus. Also, this work aimed to study the anti-hyperlipidemic activities of crackers with 100% wheat flour and crackers fortified with natural additives (thyme, carrot and hibiscus powder) on rats. The experiment was performed in 36 male rats distributed into 6 equal groups. Group (1) was kept as a normal control (fed on a basal diet), while rats of the other five groups were fed on hypercholesterolemic diet for 8 weeks to induce hypercholesterolemia. Rats of group (2) were left as a positive control and those of groups (3, 4, 5 and 6) were given unfortified cracker 100% wheat flour (CWF), 10% thyme(Cth), 10% carrot (CCa) and 10% hibiscus (Chi). The results revealed that rats consumed crackers fortified with natural additives decreases adiposity index, body mass index, Serum Cholesterol, Triglycerides (TG), Low density lipoprotein cholesterol (LDL-C) and Very low density lipoprotein cholesterol (VLDL-C), while there was an increase in serum HDL-C. Also showed decreases in serum ALT, AST, AP, Uric acid, Urea and Creatinine levels. The best results showed in thyme crackers then carrot followed by hibiscus for the treatment of hypercholesterolemic.

Keywords: Natural Additives, Crackers, Total phenols, Hyperlipidemia, Rats

INTRODUCTION

Crackers are baked food typically made from flour, flavorings such as salt, seeds, herbs and/or cheese, may be added sprinkled on top or in dough before baking Sompong, *et al.*, (2011). Some of these crackers contain elements, vitamins that are beneficial for health (Wangcharoen *et al.* 2005). However, may be crackers are harmful to health, because they have high content of oil, fat, sugar or salt which can cause dental caries, malnutrition, obesity, diabetes, coronary heart disease and may increased serum total cholesterol. (Sutharut and Sudarat, 2012).

Crackers using for its micronutrients, crackers regard as snake than a meal and therefore unlikely to replace meals given to child home and also it is easy to disrupts, no preparation, long shelf life (Asal, 2004).

Some modern plants consumed as a natural spices and herbs, containing good amounts of several nutrients, nutritional sources of minerals, vitamins, carotenoids and phenols (Guil *et al.*, 1997).

There is a worldwide to return to natural economic resources, such as thyme (*Thymus vulgaris L.*) which is widely cultivated as tea, spice and herbal medicine (Domaracky *et al.*, 2007). The leaves of thyme and its essential oil have been used in foods for flavour, aroma and preservation so added to meat, fish and food products.

Thyme uses changed to a serious drug in rational phytotherapy after a traditional herb. Thyme has various beneficial effects, as antiseptic, antimicrobial, bactericidal, anthelmintic, antioxidant properties and it has recently suggested as a natural antioxidant (Rasooli *et al.*, 2006). Thyme is an excellent source of iron, manganese and vitamin K. It is also a very good source of calcium (Sasaki *et al.*, 2005). Moreover, thyme, promotes blood circulation and functions as an exciting stimulant for the entire system.

The therapeutic effect of thyme inasmuch of its contents of flavonoids, thymol, eugenol, carvacrol, aliphatic phenols as well as saponins, tetramethoxylated flavones and luteolin. (Dorman and Deans, 2000 and Amarowicz *et al.*, 2008).

Vegetables and fruits are valuable sources of health promoting substances active in neutralization of reactive

oxygen species. Among them carrot (*Daucuscarota L.*) has nutritional value and high concentration of bioactive constituents (Maria *et al.*, 2013). Carrot is one of the most important nutritious vegetables. It is the richest source of vitamin A and β -carotene. (Deng *et al.*, 2012).

Carotenoids are precursors of vitamin A as well as excellent antioxidants which has been commonly known (Simon 1990). Consumption of carrot and its products has increased due to the recognition of antioxidant and anticancer activities of β -carotene in carrot, (Dreosti, 1993).

Hibiscus sabdariffa L., also known as roselle, it used as food and fibre. In China seeds hibiscus used for oil and plant is used as medicinal properties. Additionally, it is used in the food industries and pharmaceutical. Hibiscus has phytochemical, pharmacological and toxicological properties (Ali and Blunden, 2005) and it has an effectiveness in treatment of hypertension and hyperlipidemia effect (Hopkins, *et al.* 2013).

The Hibiscus leaves are good source of polyphenolic compounds. Neochlorogenic acid, chlorogenic acid, cryptochlorogenic acid, caffeoylshikimic acid are major identified compounds and flavonoid compounds such as quercetin, kaempferol and their derivatives. (Zhen and Jing 2016). Flowers are rich in anthocyanins, as well as protocatechuic acid. Dried flower contain flavonoids as gossypetin, hibiscetine and sabdaretine. Major pigment hibiscin which has been identified as daphniphylline. Roselle seeds are a good source of antioxidants, particularly gamma-tocopherol. (Mohamed *et al.* 2007).

The present study was carried to evaluate the possibility of enhancing antioxidant activities by using thyme, carrot and hibiscus powder added to crackers as natural antioxidants to help in industry to attract children to natural colours and its nutrition and therapeutic effect.

MATERIALS AND METHODS

Materials:

Thyme (*Thymus vulgaris L.*), Carrot and *Hibiscus sabdariffa L.* were purchased from local market for herbs and medicinal plants, Cairo, Egypt.

Soft wheat flour 72% extraction was obtained from local market in Cairo, soy flour and soy milk were obtained from soy factory.

Biochemical and durg:

Cholesterol: were obtained from El-Gomhoria Pharmaceutical Company, Cairo, Egypt.

Kits: Biochemical kits were obtained from Gama Trade Company, Dokki, Egypt.

Animals: A total of 36 male Sprague-Dawley rats (100 ±10 g) were provided from of National Research Center, Cairo, Egypt. Rats were housed as groups in wire cages under the normal laboratory conditions. The basal diet prepared according to (NRC 1995). The vitamin and mineral mixture had the prepared according to (Campbell, 1963).

Methods :

-Cracker preparation:

The unfortified cracker was prepared using 100% wheat flour (C_{WF}), other formulated using amounts of powder 10% thyme powder (C_{th}) to produce green dough, 10% carrot powder (C_{Ca}) to produce yellow dough and 10% hibiscus powder (C_{hi}) to produce red dough with 25 ml soy milk, 20 gm butter, 25 gm sugar, 2 gm salt, 3 gm baking powder, 3 ml vanilla flavor. Blend the ingredients, bake in an oven to 190 c according to the method of (Abd El Rahim *et al.* 2003).

Organoleptic evaluation of crackers:

Crackers samples were evaluated organoleptically by a panel of ten panelists for appearance, color, thickness, crispiness, shrinkage, taste and odor as the method described by Smith (1972).

Chemical analysis:

-Analytical Methods: Moisture content, protein, crude fat and ash were determined in unfortified and fortified crackers according to (A.O.A.C.1995). Total carbohydrates were calculated by difference.

Determination of antioxidant activity%: Antioxidant activity% in thyme, carrots and hibiscus powder were determined according to (A.O.A.C. 2007) at Central Lab. of Food Technolpgy Research Institute Agric. Rec. Cent., Egypt.

Determination of phenolic compounds: Phenolic compounds were determined by HPLC as gallic in thyme, carrots and hibiscus powder according to the method of (Goupy *et al.*, 1999), in Food Technolpgy Research Institute Agric. Rec. Cent., Egypt.

-Rheological properties of dough:

1- Farinograph parameters:

Water absorption and other mixing characteristics of dough prepared from control and all treatment samples were determined according to the methods described by (A.A.C.C., 2002) by using farinograph instrument (300g. of flour for each samples), at Food Technology Research Institute at Giza city, Egypt.

2- Extensograph parameters:

The Extensograph parameters is dough resistance to extension (R) (B.U(Bran bender unit), dough extensibility (E)(mm), proportional number (P.N)(R\E) and dough energy (cm²) for wheat flour and all treatment samples were measured according to procedures described in the (A.A.C.C., 2002) by using Extensograph instrument (300g. of flour for each samples), at Food Technology Research Institute at Giza city, Egypt.

Induction of obesity:

Hypercholesterolemic prepared by adding 10 % lard, 1% cholesterol and 5% bile salt to the diet fed for 8 weeks

induce a case of hypercholesterolemia as described by (Knapka and Judge 1974).

Experimental design:

Thirty six adult male albino rats Sprague Dawley strain (100 ±10 gm) obtained from the Experimental Animal House of Helwan , Egypt. All rats were allowed to free access drinking of water and basal diet for seven days adjustment to the laboratory environment. Then rats were randomly divided into 6 groups (each of 6 rats) as following:

Group (1): Negative control (ve-); rats fed on basal diet.

Group(2):Positive control (ve+); rats fed on hypercholesterolemic diet.

Groups (3): Rats fed on hypercholesterolemic diet with crackers control 100% wheat flour C_{WF} (10g/100 g /diet).

Group (4): Rats fed on hypercholesterolemic diet with fortified crackers with 10% thyme powder C_{th} (10g/100g diet).

Group (5): Rats fed on hypercholesterolemic diet with fortified crackers with 10% carrot powder C_{Ca} (10g/100g diet).

Group (6): Rats fed on hypercholesterolemic diet with fortified crackers with 10% hibiscus powder, C_{hi} (10g/100g diet).

After eight weeks experiment, body weight of rats was recorded, and rats were then euthanized by prolonged exposure to ether anesthetic. The abdomen was opened, and body fats, including mesenteric, visceral, epididymal and retroperitoneal fats were carefully dissected out and total fat mass was weighed. The adiposity index (Ad. I) was calculated by dividing total body fat mass by body weight and multiplied by 100 (Ad. I = fat weight [F.wt]/Final B.wt × 100) as described by (Pichon *et al.*, 2006).

Rats were weighed in grams and lengths were measured in cm at the end of eight weeks study. The body mass index (BMI) was calculated (by dividing the body weight in kilograms by the length in meters squared) (Buettner *et al.*, 2006). The body weight and body length were measured and used to determine the following parameters:

Body Mass Index (BMI) = body weight (g) / length² (cm)

At the end of the experiment period, the rats were fasted overnight then the rats were anaesthetized and sacrificed and blood samples were collected from the aorta. The blood samples were centrifuged and the serum was carefully separated into dry clean Wassermann tubes by using a Pasteur pipette and kept frozen till analysis at -20°C.

Biochemical analysis of serum:

Determination of lipids paeameters were determined by enzymatic methods as follows: Total cholesterol (TC), high-density lipoprotein cholesterol (HDL-c) and triglycerides (TG)), according to (Richmod, 1973, Lopes-Virella,*et al.*, 1977, and Fossati and Prenape, 1982). While (LDL-c and VLDL-c) were calculated according to the equation of (Friedwald *et al.*, 1972). Atherogenic Index (Total cholesterol / HDL-cholesterol) were calculated according to the equation of (Golay *et al.* , 1990).

Uric acid was determined in the serum according to the method described by Fossati *et al.*, (1980). Urea nitrogen was determined according to Patton and Crouch, (1977). Creatinine was determined according to Bartels *et al.*, (1972). Serum alanine and aspartae aminotransferase (ALT, AST) , alkaline phosphates (AP) enzymes, were estimated according to Reitman

and Frankel (1957), Kind and King (1954) and Weichselbaum (1946) respectively.

Determination of serum antioxidant parameters

Total Antioxidants Capacity (TAC), Superoxide Dismutase (SOD) activity, and Malondialdehyde (MDA) were determined according to Nishikimi *et al.*, (1972); Cao *et al.*, (1993) and Ohkawa *et al.*,(1979), respectively. While Acetylcholine esterase (AChE) activity was determined colorimetrically according to Hestrin (1949).

Statistical analysis:

The obtained data were statistically analyzed using computerized SPSS (Statistic Program Sigmatat, Statistical Soft-Ware, SAS Institute, Cary, NC). Effects of different treatments were analyzed by one way ANOVA (Analysis of variance) test using Duncan’s multiple range test and $p < 0.05$ was used to indicate significance between different groups (Snedecor and Cochran, 1967).

Table 1. The organoleptic properties of unfortified crackers made from wheat flour and fortified crackers with thyme, carrot and hibiscus powder.

Samples of biscuits	Appearance (10)	Color (15)	Thickness (15)	Crispness (15)	Shrinkage (15)	Taste (15)	Odor (15)	Total (100)
unfortified	9	14	13	14	14	14	14.5	92.5
Fortified crackers with10% Thyme	9	13	13	14	14	13.5	13.9	90.4
Fortified crackers with10% Carrot	9	14	13	14	14	14.7	14.2	92.9
Fortified crackers with10% Hibiscus	9	14	13	14	14	14.7	14.5	93.2

W.F = Wheat flour

Chemical composition of crackers made from wheat flour CWF and fortified crackers with thyme, carrot and hibiscus powder:

Data in Table (2) showed the gross chemical composition of crackers processed from wheat flour 72% and thyme, carrot and hibiscus powder. Results indicated that moisture content was 8.0 % in crackers formulae control. All crackers formulae were higher in moisture content. Results also, indicated that protein content of control formula was higher than those of the

RESULTS AND DISCUSSION

The organoleptic properties of crackers made from wheat flour and fortified crackers with thyme, carrot and hibiscus powder:

Crackers supplemented by 10% thyme, carrot and hibiscus powder were sensory evaluated with different parameters appearance, color, thickness, crispness, shrinkage, taste, odor and overall acceptability values and compared with crackers from wheat flour as presented in Table (1). Formula contained 10% thyme, carrot and hibiscus powder showed acceptability with ability value 90.4, 92.9 and 93.2% respectively. Abdel Azim (2007) reported that natural plants have been widely used in many food products as crackers, biscuits and cookies which has great economic importance and often stored for extended periods before consumption.

other mixtures. This could be due to that the ratio of additives namely thyme, carrot and hibiscus powder contained low amount of protein in comparing with wheat flour. The ash content in crackers formula with thyme, carrot and hibiscus powder was increased slightly comparing with crackers with wheat flour only. These results are in agreement with those given by (Abd El Rahim *et al.*2003) Who indicated that crackers was higher score in fiber due to high content of its additives.

Table 2. Chemical composition of crackers unfortified and fortified with thyme, carrot and hibiscus powder.

Samples of crackers	Moisture %	Crude protein %	Crude fat %	Ash %	Carb. %
unfortified	8.00	10.53	12.33	2.0	67.14
Fortified crackers with10% Thyme	8.50	8.77	13.33	2.50	66.9
Fortified crackers with10% Carrot	9.8	8.51	13.00	2.41	66.29
Fortified crackers with10% Hibiscus	8.70	8.0	13.4	2.9	67.0

*Carb=carbohydrate

Total phenols and antioxidant activity contents of natural additives used in crackers formulation:

Total phenols and antioxidant activity % content of thyme, carrot and hibiscus (on dry weight basis) were shown in Table (3). It could be noticed from the results that thyme powder had the highest level of total phenols (12366 mg/100g), followed by carrot powder which was (5087.4 mg/100gm) and hibiscus has (4262.9 mg/100g) while antioxidant activity were 96.7%, 89.59%and 86.17% respectively for thyme, carrot and hibiscus. These result of carrot at accordance with (Ally 2001).

Table 3. Total phenolic content (mg/100g) and antioxidant activity (DPPH) radical % of natural additives used in crackers formulation.

Samples	Total phenolic Content (mg/100g)	Antioxidant activity radical (DPPH) %
Thyme	12366.0	96.7
Carrot	5087.4	89.59
Hibiscus	4262.94	86.17

Phenolic compounds of natural additives:

Polyphenolic compounds are very important constituents in activating lipid free radical chains and preventing hydroperoxide. Data in Table (4) showed that the main phenolic acids identified in thyme powder was Salicylic acid (177.7), while carrot powder had high contents of Chlorogenic, Catechol 299.72, 108.3 respectively. While hibiscus contained P-oH-Benzoic, benzoic, Epicatechen, Salicylic and Ellagic their values were 1260.7, 1200, 870.3, 625.5 and 540.01 respectively.

Carrot contains phenolic constituents as chlorogenic acid (Arscott and Tanumihardjo, 2010). Carrots contained mainly hydroxyl cinnamic acid 42.2%, chlorogenic acid represented 61.8 % of total phenolics (Zhang and Hamauzu, 2004). Chlorogenic acid, caffeic acid, *p*-hydroxybenzoic acid, ferulic acid and other cinnamic acid isomers predominated in carrots of different colors (Sun *et al.*, 2009) .

Table 4. Phenolic compounds of natural additives.

Phenolic compounds	Thyme	Carrot	Hibiscus
Pyrogallol	7.7	----	177.76
4-Aminobenzoic	-----	-----	15.52
Chlorogenic	13.78	-----	225.68
P-oH-Benzoic	12.29	-----	1260.70
Epicatechen	7.06	-----	870.39
Caffeic	5.99	29.35	70.30
Vanillic	2.77	17.97	69.74
Ferulic	5.89	15.65	50.82
Benzoic	-----	80.24	1200.08
Salicylic	177.7	-----	625.52
Coumarin	7.001	-----	160.82
Ellagic	16.95	17.70	540.01
Cinnamic	8.006	-----	26.40
Gallic	0.52	15.77	-----
Protocatechuic	13.89	6.44	-----
Syring	-----	-----	-----
Catechol	-----	108.36	-----
Caffeine	-----	7.71	-----
Chlorogenic	-----	299.72	-----

Anthocyanins found in hibiscus are delphinidin-3-sambubioside, delphinidin-3- glucose cyaniding and 3 sambubioside. (Christian *et al.* 2006). Hibiscus also contains alkaloids, L- ascorbic acid , citric acid, anisaldehyd, β -carotene, gossypetin, hibiscetin, β -sitosterol, galactose and mucopolysaccharides (Hirunpanich, *et al.* 2005).

Free radicals can be destroyed by antioxidants present in the body so that the biological damage by phenolic compounds can be avoided (Vincent *et al.*2004).

The water extract of dried hibiscus showed the presence of catechin (4.25%) and ellagic acid (28.20%) (Lin *et al.*, 2012), while (Yang *et al.*, 2010) found that hibiscus contained gallic acid (27.98%) protocatechuic acid (24.24%), caffeic acid (19.85%), catechin (2.67%) and gallic acid (2.44%).

Rheological properties of dough: Farinograph parameters

The results in Table (5) showed that, the flour control absorbed 60.6% of water, when the flour with carrot absorbed 69.4% of water, hibiscus absorbed 63.3% then thyme absorbed 61.23%, These result indicated to increase in absorption of water when added natural additives because its high content of fiber. These results agreed with (Unver and Domolds 1976) who found that increase in water absorption of dough when protein content decreased. Table (5), showed that, flour control had arrival time 1.5 and flour with natural additives increased in arrival time which ranged from 1.7 to 2.3 min. also Table (5), showed that, dough development in flour control equal 1.0 min, when flour with natural additives had dough development more than flour control which equal 1.5 min. At these study value of dough stability for flour control were 1.0 min which indicated to weak dough, when value of dough stability for flour with natural additives were 2.0 min. for thyme dough, 3.5 for carrot dough and 1.5 for hibiscus dough. These results indicate to the flour with natural additive powders had the ability barrier to gas which effects on the freshness of the product. Table (5), showed that, the degree of softening of flour control were 70 B.U, when the degree of softening of flour with thyme and hibiscus powder were 80 B.U, while the degree of softening of flour with carrot powder were 90 B.U these mean that flour with natural additives had the heights degree of softening, these results indicated to the addition powders to flour increase the degree of softening of dough, This result confirms the previous result for dough stability. From these results we can conclude that, the fortified products with thyme, carrot and hibiscus will have lowest staling and lowest firmness than control products, because the high softening led to produce product with antistaling and low firmness. (Dapčević *et al.* 2009) reported that, degree of softening is predominantly influenced, as it was for the dough stability, by the amount and quality of gluten, these result agree with the result at above.

Table 5. Farinograph parameters of dough formulae from wheat flour and samples with some herbs.

Samples	Water absorption %	Arrival time (min)	Dough development (min)	Dough stability (min)	Degree of softening (B.U)
Unfortified flour	60.60	1.5	1.0	1.0	70
Fortified flour with10% Thyme	61.23	1.7	1.5	2.0	80
Fortified flour with10% Carrot	69.40	2.3	1.5	3.5	90
Fortified flour with10% Hibiscus	63.3	1.9	1.5	1.5	80

Data in Table (6), showed the effect of natural additives on dough elasticity or dough resistance (R), elasticity or resistance of dough from flour control are higher than the elasticity of dough from flour with additives because the elasticity of dough from flour control equal 510 B.U, but the dough from flour with thyme, carrot and hibiscus were 380, 480 and 350 B.U respectively, also Table (6), showed the extensibility of dough control and dough from flour with natural additives, the dough which made from flour with thyme, carrot and hibiscus had a lower value of extensibility than dough control which had 195 mm, Table (6), showed the value of P.N for dough with or without any addition, this result indicated to the

flour control had P.N equal 2.6, while thyme and carrot powder dough had 3.6 and 4.5P.N respectively, then the flour with hibiscus had 2.5 P.N. Value of energy for dough which made from flour control was 85 cm², then the value of flour with carrot powder had higher value was 128 cm². These results agreed with (Gurpreet *et al.* 2014) who found that Water absorption increased for all spices and herbs; cinnamon, clove and thyme except for garlic powder. Dough development time was increased by adding cinnamon, clove, garlic and thyme. Incorporation of cinnamon, thyme and clove increased the paste temperature due to high fibre content, so gluten was competing with fibre present in spices and herbs for water.

Table 6. Extensograph parameters of dough formulae from wheat flour and samples with some herbs.

Samples	Extensibility (mm)	Elasticity (B.U)	proportional number (P.N)	Energy (cm2)
Unfortified flour	195	510	2.6	85
Fortified flour with10% Thyme	105	380	3.6	67
Fortified flour with10% Carrot	105	480	4.5	128
Fortified flour with10% Hibiscus	140	350	2.5	80

Effect of prepared cracker control and with herbs on fat weight (F.wt), body weight gain ratio, adiposity index (Ad.I) and BMI of hypercholesterolemic rats:

The untreated cholesterol group (positive control) feeding on hypercholesterolemic diet for 8 weeks showed significant increase in fats weight (F.wt), body weight gain, Ad.I and body mass index (BMI) when compared to negative control rats in Table 7. While treated rat groups which feed on unfortified and fortified crackers C_{WF} , C_{th} ,

C_{Ca} and C_{hi} (100% wheat flour, 10% thyme, 10% carrot and 10% hibiscus powder) given to obese rats for 4 weeks induced significant decrease in fats weight (F.wt), weight gain and AdI at compared with positive control group as shown in Table 7. These results agree with those reported by (Reddy *et al.* 2005) who reported that natural phenolic compounds added to crackers were widely used to retard lipid oxidation, any ways of adding these compounds into products as ingredients.

Table 7. Effect of prepared crackers control and with some herbs, on fat weight (F.wt), body weight gain ratio, adiposity index (Ad.I) and BMI of control and hypercholesterolemia rats.

Parameters	Groups	F. wt g/ day	B.W.G. g/ day	Ad.I (%)	BMI (gm)/(cm)2
Negative Control		5.48±0.15c	45.19 ± 4.28b	3.5±0.10d	0.37
Positive Control		14.50±0.22 a	98.95 ± 7.89 a	6.93±0.15a	0.41
Unfortified crackers		7.44±0.13 b	41.65 ± 5.89c	4.90±0.12c	0.38
Fortified crackers with10% Thyme		7.00±0.10c	43.56 ± 3.21bc	4.55±0.18c	0.39
Fortified crackers with10% Carrot		6.71±0.10c	45.01 ± 4.26b	4.32±0.15c	0.40
Fortified crackers with10% Hibiscus		7.13±0.13 b	42.56 ± 4.21bc	4.67±0.10c	0.38

Values with the same letters indicate non- significant difference (P<0.05) and vice versa.

Effect of prepared cracker control and with herbs on lipids profile and atherogenic-index of hypercholesterolemia rats:

As demonstrated in Table 8, rats feeding on hypercholesterolemic diet for 8 weeks produced significant (P < 0.05) increases in serum levels of TC, TG, LDL-c and VLDL-c in positive control when compared with normal control rat group. However there was a significant decrease in TC, TG, LDL-c , VLDL-c and atherogenic index levels of treated rat groups unfortified and fortified crackers with (100% wheat flour, 10% thyme, 10% carrot, and 10% hibiscus powders) when compared with untreated cholesterol rats (positive control) group (p<0.05). However there is decrease between the treated groups fortified

crackers with C_{th} , C_{Ca} and C_{hi} (10% thyme, 10% carrot, and 10% hibiscus powder,), the best reduction in cholesterol level, atherogenic index in thyme group. The results were in the line with (Ozkol *et al.* 2013) who found that the lipid profile was ameliorated especially by supplementations of thymus vulgaris on diabetic rats. This results also agree with (Hopkins *et al.* 2013) who study the effectiveness of hibiscus in the treatment of hypertension and hyperlipidemia. And (Manach *et al.*, 2004) who found the relationship between the antioxidant compounds in plants and their effectiveness in the treatment of these diseases have been previously described.

Table 8. Effect of prepared crackers control and with some herbs powder on lipids profile and atherogenic index of control and hypercholesterolemia rats.

Parameters	Groups	T. Ch mg/dl	T.G. mg/dl	HDL-C mg/dl	LDL-C mg/dl	VLDL-C mg/dl	Atherogenic Index
Negative Control		77.30±3.66c	81.84±3.65b	41.75± 1.13a	19.18±1.23c	16.37±1.08b	1.85±0.098b
Positive Control		156.69±4.98a	166.58±3.69a	31.48±1.87b	59.89±5.67a	33.32±1.82a	4.98±0.076a
Unfortified crackers		80.28±4.45c	93.46±2.55b	39.12±2.11a	22.47±2.01c	18.69±1.64b	2.05±0.056b
Fortified crackers with10% Thyme		75.19±2.84c	89.19±5.23b	42.24±1.09a	15.11±2.17c	17.84±2.09b	1.78±0.091b
Fortified crackers with10% Carrot		77.19±2.19c	82.46±4.54b	41.55±1.54a	19.15±2.44c	16.49±1.33b	1.8±0.056b
Fortified crackers with10% Hibiscus		79.67±1.99c	80.52±1.96b	43.61±2.03a	19.96±3.22c	16.10±1.11b	1.83±0.054b

Values with the same letters indicate non- significant difference (P<0.05) and vice versa.

Effect of prepared cracker control and with herbs on serum kidney function parameters of hypercholesterolemia rats:

On the other hand, the study also showed some kidney function parameters of different treated rat groups (Table 9) the uric acid, urea and creatinine levels of untreated rat group (positive control) were significantly different (p<0.05) from the normal control rat group. However treated rat group C_{WF} feeding on unfortified crackers significantly reduced uric acid, urea and creatinine (p<0.05) compared to positive control, while treated rat groups with fortified crackers C_{th} , C_{Ca} and C_{hi} improved these parameters to a level similar compared to normal control rat group. The highest levels of uric acid, urea nitrogen and creatinine levels in untreated cholesterol group (positive control) were 3.87, 25.48 and 2.75 respectively while rats feeding on crackers with C_{WF} (100% wheat flour) were 2.01, 18.95 and 1.49, respectively. Rats feed on crackers fortified with thyme showed the best results in urea and creatinine levels. These results agreed with (Amarowicz *et al.*, 2008) who reported

that therapeutic potential of thyme depended on its contents of flavonoids, thymol, eugenol, carvacrol, aliphatic phenols as well as luteolin, tetramethoxylated flavones and saponins.

Effect of prepared cracker control and with herbs on some liver functions of control and hypercholesterolemia rats:

The data in Table (10) showed some liver function parameters, it was observed that the untreated cholesterol rats group (positive control) showed significant increase in ALT, AST and ALP at p <0.05 in comparing with normal control group. The treated rats groups C_{th} , C_{Ca} and C_{hi} had the lowest ALT, AST and ALP levels, the best values were C_{th} group 30.71, 52.37 and 45.10 respectively while C_{Ca} group were 32.84 , 52.11 and 45.91 respectively followed by C_{hi} which were 34.11, 57.71 and 47.34 respectively. (Monira and Naima 2012) concoulded that treatment with thyme extract in paracetamol intoxicated rats reduced levels of serum transaminases, ALP and total bilirubin towards normal indicating to its hepatoprotective effect and demonstrated membrane stabilizing activity of thyme extract.

Table 9. Effect of prepared cracker with some herbs on of serum kidney function parameters of control and hypercholesterolemia rats.

Parameters	Groups	Uric acid mg/dl	Urea μ /mg	Creatinine mg/dl
Negative Control		1.73±0.26c	19.75± 1.13a	1.27±0.01 b
Positive Control		3.87±1.01 a	25.48±1.87b	2.75±0.11 a
Unfortified crackers		2.01±0.81b	18.95±1.54a	1.49±0.02 b
Fortified crackers with10% Thyme		1.91±0.77 c	15.62±2.11a	1.68±0.12 b
Fortified crackers with10% Carrot		1.97±0.67 b	17.21±2.03a	1.75±0.13 b
Fortified crackers with10% Hibiscus		1.84±0.74 c	18.24±1.09a	1.18±0.18 b

Values with the same letters indicate non- significant difference (P<0.05) and vice versa.

Table 10. Effect of prepared cracker with some herbs on some liver functions of control and hypercholesterolemia rats.

Parameters	Groups	ALT μ /ml	AST μ /ml	ALP μ /ml
Negative Control		29.35±1.12b	51.17±5.81c	44.17±5.66 b
Positive Control		46.55±3.35 a	79.39±9.61 a	60.38±5.81 a
Unfortified crackers		36.17±2.01 b	55.14±8.10c	49.73±4.37 b
Fortified crackers with10% Thyme		30.71±1.81 b	52.37±6.01c	45.10±4.11b
Fortified crackers with10% Carrot		32.84±0.74 b	52.11±4.13 c	45.91±3.11 b
Fortified crackers with10% Hibiscus		34.11±3.65 b	57.71±6.15 c	47.34±5.01 b

Values with the same letters indicate nonsignificant difference (P<0.05) and vice versa.

AST: Aspartate amino transferase ALT :Alanine amino transferase ALP: Alkaline phosphatase

Effect of prepared cracker control and with herbs on Serum Total Antioxidant Capacity (TAC), Malondialdehyde (MDA), Superoxide Dismutase (SOD), and Acetylcholine Esterase (AChE) of control and hypercholesterolemia rats.

The result postulated in Table (11) revealed that levels of TAC and SOD for normal control rats were (3.99 ±0.58 u/mg and 30.97±0.54 μ /ml), while the corresponding levels for positive control group were lower (1.12±0.35 u/mg and 20.54 ±0.71 μ /ml). Data showed a significant increase in TAC and SOD levels and a significant decrease in MDA in all rat groups which treated with fortified cracker with herbs as compared to the positive control groups. The treated rats groups with C_{th}, C_{ca} and C_{hi} feeding on fortified cracker increased total antioxidant capacity TAC and SOD

and decreased MDA levels comparing with positive control. The treated rat group C_{th}, C_{ca} and C_{hi} feeding on fortified cracker had the highest acetylcholine esterase (AChE) levels which was similar to the values of normal control group. Fortification with herbs to diet increased antioxidant parameters such as TAC , SOD, and acetylcholine esterase (AChE) levels. These results agreed with (Rasooli *et al.*, 2006) who reported that thyme possess various has beneficial effects, as antimicrobial, antiseptic, antioxidant properties and it has recently suggested as a natural replacement for synthetic antioxidant. (Atawodi, 2005) studied that, hibiscus showed tremendous promise for preventive intervention in the pathogenesis of many diseases, such as cancer and ulcer this rested to the antioxidant compounds in plants

Table 11. Effect of prepared cracker with some herbs on serum total antioxidant capacity (TAC) malondialdehyde (MDA), superoxide dismutase (SOD), and acetylcholine esterase (AChE) of control and hypercholesterolemia rats.

Parameters	Groups	TAC (U/mg)	MDA μmol/mg	SOD (μ /ml)	Acetylcholine esterase (AChE) Nmol
Negative Control		3.99±0.58a	1.54±0.54d	30.97±0.54a	7.15± 0.4 ^a
Positive Control		1.12±0.35d	4.79±0.65a	20.54±0.71d	3.54± 0.4 ^c
Unfortified crackers		2.42±0.77c	2.11±0.58 bc	24.52±0.28c	5.96± 0.7 ^a
Fortified crackers with10% Thyme		2.99±0.68 b	2.13±0.35d	26.98±0.84ab	6.15± 0.6 ^a
Fortified crackers with10% Carrot		2.34±0.66 b	2.68±0.32b	25.85±0.25 b	5.89± 0.4 ^b
Fortified crackers with10% Hibiscus		2.17±0.98b	2.64±0.74d	24.87±0.24 b	4.19± 0.4 ^b

Values with the same letters indicate non- significant difference (P<0.05) and vice versa.

CONCLUSION

From The results obtained, it could be underline the beneficial effect of using natural antioxidant for stabilizing in crackers, its phenolic compounds which are one of the most important groups and its effect on hypercholesterolemic rats.

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