Effect of Replacing Soybean Concentrate with *Bulgur* (Wheat Groats) Flour on Quality and Storage Stability of Beef Burger

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ABSTRACT

Replacing soybean concentrate with *Bulgur* flour in beef burger manufacture was studied. The results showed that the components of *Bulgur* were carbohydrate (82.65 %), protein (13.47%), moisture (11.59%) and fat (1.8%). Dietary fiber content was 31.33 % NDF and 1.98% ADF. Water absorption, fat absorption and emulsion stability were 173.74%, 175.87% and 39/100g, respectively. Soybean concentrate had high level of isoflavones comparing with *Bulgur* flour. Genistein , biochanin A, daidzien and formononetin in soybean concentrate were higher 13, 6, 12 and 8 times than that in *Bulgur* flour. Percentage of DPPH radical scavenging activity in *Bulgur* and soybean was 28.84% and 54.42%, respectively. Five different formulations of beef burger were prepared, 3 containing 10%, 15% and 20% *Bulgur* flour , one having 20% soybean concentrate and the last one was 100% meat .The moisture, protein, fat, ash and carbohydrate contents ranged from 66.37 to 72.72, 59.13 to 70.86, 21.48 to 24.34, 3.78 to 4.81 and 0. 17 to15.48%, respectively in the beef burger samples. Cooking loss and shrinkage increased after 3 months of freezing at –20°C, meanwhile decreasing in WHC was traced. The TBA values of the different burger products and after storage 3 months at –20°C ranged from 0.008 to 0.018 mg malonaldehyde per 1 kg sample. The TVC ranged from 3.5×10^3 to 6.0×10^4 CFU/g before and after frozen storage. Enterobacteriaceae was 7.2×10^2 to 5.1×10^3 CFU/g in samples at zero time and not detected after one month of storage at –20°C. *E.coli*, *Coliform* and *Staphylococcus aureus* were not detected in all samples before and after 3 months of frozen storage . All roasted samples were judged acceptable by the panelists, 10% and 15% beef burger with *Bulgur* had the highest acceptability.

Key words: Bulgur, soybean, beef burger, isoflavonesd.

INTRODUCTION

Meat plays a pivotal role in alleviating malnutrition due to its content of high quality protein, vitamins, minerals and fat (Mallika *et al.*, 2009). The main problem of meat and meat products is the perishability due to their very low inherent antioxidant capacity (Gupta & Savalia, 2012). The antioxidants prevent fat oxidation, reduce the harmful free radicals and protect cells from damage. (Ruban *et al*., 2009) .The antioxidant capacity of meat and meat products can be improved by natural antioxidants such as flavonoids present in different plant sources such as legumes, cereal, vegetables ,fruits and their industrial by-products (Hue *et al*., 2011). These materials are also rich in dietary fibers. Therefore, the incorporation of such sources at suitable levels enhances the cooking yield, emulsion stability and functional value of meat products. (Perumalla *et al*., 2011).

Flavonoids are phenolic compounds having antioxidants and chelating properties. They form from three major classes, flavones (such as, quercetin and rutins), isoflavonoids (such as, genistein, glycitein, daidzein, formononetin) and neoflavonoids (Tapas *et al*., 2008). The antioxidant activity of these compounds is mainly due to high reactivity of their hydroxyl groups .The recommended dietary allowance of flavonoids is not fixed (Kumar *et al*., 2013). The USDA (2011) stated that for getting the therapeutic and health promoting benefits ,a minimum 50-500 mg of flavonoid per day are recommended. Acidity and processing of food (boiling and cooking) may cause up to 50% destruction of flavonoids (kumar *et al*., 2013). In India, the average daily intake of flavonols is about 1-2 g per day out of which the mean intake of flavonols and flavones accounts about 23mg/day and quercetin about 16 mg /day. For rat, the LD_{50} is 2-10 g per animal for most flavonoides. Such level is unrealis-

tic in normal diet (Sandhar *et al.,* 2011). Kumar *et al*. (2013) suggested daily intake of less than 1 mg of flavonoids per adult per day for human- being. Generally excess intake of flavonoids causes diarrnoea due to problem in digestion.

Beef burger is one of the meat popular product overall the world. The meat manufacturers continually try to improve processing quality, storage stability, safety, sensory attributes, nutritive value and functionality of their products .Such properties can be achieved by replacing part of meat by some types of plant sources rich in phenolic compounds such as flavonoids and dietary fibers. Soybean is one such sources. It is rich in isoflavones, a one class of flavonoids. It was used as meat replacer during preparing beef burger .Soybean flavonoids have some health benefits such as protection against breast and prostate cancers ,heart disease and osteoporosis (Hendrich *et al*. 1994) .Recently, the similarity between chemical composition of isoflavones and esterogen hormone causes controversy about the consumption and utilization of soybean in food production (Kumar *et al*., 2013). Also, isoflavones devoid of reactive hydroxyl groups and sequentially showed little antioxidant activity. According to Cai *et al*. (2006), the antioxidant capacity of flavonoids having the following order as flavanols> flavonols> chalcones> flavones> isoflavone.

Bulgur (wheat groats) is rich in dietary fibers, vitamin B$_1$, B$_2$ and phenolic compounds .Generally *Bulgur* is a staple for centuries in many Middle and Near Eastern countries due to its low cost, long storability, simple processing, high nutritional value (Köksel *et al*., 1999) and health protection effect against cardiovascular and heart diseases (Ozvural & Vural , 2008). *Bulgur* is commercially manufactured from soaked, blanched, gelatinized and dried durum wheat grains (Bayram, 2007).

Therefore the present study is aimed to replace soybean concentrate with *Bulgur* flour in beef burger manufacture. The influence of such replacement on quality and storage stability of this product was investigated.

MATERIALS AND METHODS

Materials

Brazial frozen imported back rip boneless beef, beef tallow ,fresh onion, refined fine iodized common salt, spices blend mixture of National Co., corn oil, *Bulgur* (wheat groats) of Al Suhagy for Food Industries, soybean concentrate of Bakri Packing Co., grease-proof paper, polyethylene bags and foam plates (22×17cm) were purchased from Alexandria local market, Egypt.

The following standards isoflavone : genistein (4′,5,7,trihydroxy isoflavone), daidzein (4′,7, dihydroxyisoflavone), formononetin (7-hydroxy -4′- methoxyisoflavone) and biochanin A (5,7-dihydroxy -4-methoxy isoflavone) were obtained from Sigma –Aldrich ,USA. All solvents used in extraction and determination were of high –purity "HPLC" grade (Merck, Darmstadt, Germany) ,other reagents and chemicals were analytical grades.

Methods

Technological methods:

Beef burger preparation:- Frozen meat was thawed at room temperature (22±3°C) for 4-5 hr, dressed by removing their surrounded fat layers, cut into 10 cm thickness portions then mixed with beef tallow at 9:1 (w/w) ratio and minced 2 times through 5mm plate of Luska meat chopper .To the resultant minced meat ,4% of small pieces of fresh onions,1% salt, 1% spices mixture, and 20% cold water were added and well mixed. Five different formulations of beef burger were prepared, 3 containing 10%, 15% and 20% *Bulgur* flour ,one having 20% soybean concentrate and the last one was 100% meat as the control. Each formula was chopped 2 times using Luska chopper, formed into around pieces with 10 cm diameter, 1cm thickness and 70g weight. Each piece was surrounded with 2 pieces of grease-proof paper, then packed in polyethylene bags. The bags were kept at -20°C in deep freezer (Aristone) for 3 months.

Cooking of beef burger :- Raw burger samples were roasted in roasting pan at 140°C until center temperature reached 80°C ,then cooled to room temperature (22±3°C).The cooking loss and shrinkage were calculated using the following equations as mentioned by El –Magoli *et al*. (1996).

$$\text{Cooking loss (\%)} = \frac{w_1 - w_2 \times 100}{w_1}$$

Where:-

W_1: weight of beef burger sample before cooking.

W_2: weight of beef burger sample after cooking.

$$Shrinkage\ (\%) = \frac{A_1 - A_2 \times 100}{A_1}$$

Where:

A_1 = area of burger sample before cooking.

A_2 = area of burger sample after cooking.

Analytical method:-

Physical properties:-

Water holding capacity (WHC) of beef burger was determined using filter paper press method (Aman, 1983).Water and fat absorption of *Bulgur* flour were determined according to Del Rosario & Flores (1981) using distilled water and corn oil ,respectively. The emulsion capacity was estimated by formation suspension of *Bulgur* flour, distilled water and corn oil, then homogenized ,heated at 80°C,cooled and centrifuged as stated by Chau & Cheung(1998).

The colour values, lightness (L*) ,redness(a*) and yellowness(b*) of burger samples were evaluated using a Hunter Lab Ultra Scan VIS model, colorimeter (USA).The instrument was standardized during each sample measurement with a black and white tail (L*= 94.1 ,a*= 1.12, b* =1.26). Five reading of each colour index of Hunter scale (L*, a*,b*) were recorded (Santipanichwing & Suphantharika,2007).

Texture properties were analyzed by texture profile analysis (TPA) in a TA-XT plus Texture Analyzer (Texture Pro CT3 V1.2,Brookfield, Middleboro, USA) as described by Yuan & Chang (2007) .Burger section (height-20 mm) were axially compressed to 40% of their original height. Force time deformation was obtained with 10 kg load cell, applied at a cross head speed of 1mm/s. Attributes calculated were hardness, cohesiveness, and chewiness.

Chemical analysis:-

Proximate composition : Moisture, protein, fat and ash contents of beef burger and *Bulgur* flour were determined according to the AOAC (2000) carbohydrate content was calculated by difference .Thiobarbituric acid (TBA) was calorimetrically estimated according to Park *et al*. (2007) using UV-VIS Spectrophotometer Laxo alpha 1102, suit and expressed as mg malonaldehyde per kilogram fat or sample. Dietary fibers were determined according to the method of AOAC (1990).The pH was determined using pH meter type MVX100 Beckman

(USA) at room temperature (22 ± 3°C) as described in AOAC (2000).

Isoflavones identification by HPLC:-

Separation, identification and determination of *Bulgur* flour and soybean concentrate isoflavones were carried out as follow: Extraction of isoflavones was done as described by Wang & Murphy (1994) by stirring 2 g of sample with 10 ml of acetonitrile and 2 ml of 0.1 N HCL in 125 ml screw tap Erlenmeyer flask for 2 hr at room temperature. After filtration through Whatman No .42 filter paper, the filtrate was dried on a rotary evaporator at 30°C.The dried material was redissolved in 10 ml of 80% HPLC grade MeOH in water. Aliquot of sample was filtered through a 0.45 μm poly tetra-fluoro ethylene (PTFE) filter unit and analyzed by HPLC according to Frank *et al*. (1994) with slight modification.A unit of HPLC (Shinaezu, Japan) connected with a UV-VIS diode –array detector (Dionex –Ulti Mate 3000) and with a reversed –phase C18 (ODS) column (250x4.5 mm. Dionex – Ulti Mate 3000) was used. A volume of 25 μl sample or standards was injected at column temperature setting at 35°C. Isoflavones were separated with two mobile phases: phase A (0.1% glacial acetic acid in water) and phase B (0.1% glacial acetic acid in acetonitrile).The gradient profile was: 20% A-45% B in 20 min., 45%A- 100% B in 1 min., 100% B for 4.5 min.,100% B -20% A in 0.5 min. The flow rate was set at 0.7 ml / min. UV spectra were recorded (from 200 to 350 nm) and area responses were integrated by water PDA software .The total processing time was 20 min .The peaks of the separated isoflavones were compared with those of standard isoflavones (genistein (4′,5,7,trihydroxyisoflavone), daidzein (4′,7,dihydroxy isoflavone), formononetin (7-hydroxy-4′- methoxyisoflavone) and biochanin A (5,7-dihydroxy -4′-methoxyisoflavone). The concentration of isoflavones in the samples was calculated in terms of mg/g dry matter.

Antioxidant activity:-

Radical scavenging activity of *Bulgur* flour and soybean concentrate extracts was measured using the stable radical DPPH (2,2 diphenyl-1-picrylhydrazyl) according to Brand –Williams *et al* .(1995). The percentage of DPPH scavenging for extracts was calculated as follows:-

Radical Scavenging activity % =
$$[(A_{Control} - A_{Sample}) / A_{Control}] \times 100$$

Microbiological methods: Ten grams of beef burger were blended with 90 ml of sterilized peptone water for 2 min, in sterilized glass jar of blender. Appropriate dilution was prepared for enumeration using standard microbiological pour plate technique and the recommended culture media of Oxiod (2002). Plate count agar medium was used for enumerating the Total Viable Count (TVC), after incubating at 35-37°C for 48 hr Violet red bile agar with methyl umberliferyl glucourniod (VRB-MUG) selective media was used to isolate coliform, gram negative enteric bacteria and rapid detection of *E.coli*. The proper dilution of the beef burger homogenate was inoculated in steril petri dishes then medium was poured and plates were incubated at 37°C .for 18-24 hr Colonies of lactose negative enterobacteriaceae are colourless and those of lactose positive are red and often surround by a forbid zone due to precipitation of bile acid are coliform, light blue fluorescent colonies under UV-lamp (336 nm.) denote as *E.coli*. The recommended Difico Baried Parker agar medium by ICMSF (1978) was used to detect *Staphylococcus aureus* after incubating the plates at 35-37 °C for 48 hr the black shiny colonies with narrow white margin and surrounded by clear zones were counted as *Staphylococcus aureus*.

Sensory Evaluation : - Colour ,texture, ,taste, odour and overall acceptability of cooked beef burger were organoleptically evaluated using 10 trained panelists from, Food Science and Technology Department, Faculty of Agriculture, Alexandria University. They were asked to rate their acceptabilities of cooked burgers products according to nine point scale, ranging from the like extreme 9 to dislike extreme 1 point as described by Meilgaard *et al* .(1999).

Statistical Analysis:- Statistical analysis system (SAS) software 917 SAS (USA,2003) using two- way analysis of variance (ANOVA) was followed. The differences among means were determined for significance at $P<0.05$ using Duncan's multiple range test.

RESULTS AND DISCUSSION

Proximate composition, dietary fibers and functional properties of *Bulgur* flour: - According to the data in Table (1) the higher component of *Bulgur* is carbohydrate (82.65 %), while fat was the lowest (1.8%). *Bulgur* has a considerable amount of protein (13.74%) consisted of balance of amino acids (Wijngaard & Arendt, 2006).The results of

Köksel *et al*.(1999) showed that protein and ash contents of *Bulgur* ranged from 9.5% to 11.4% and from 1.2% to 1.95% ,respectively. Drying process of *Bulgur* can be done naturally or in towers to lower *Bulgur* moisture content to ~12% (Bayram, 2007).In the present study, moisture content of *Bulgur* was 11.59%.

Comparing with soybean protein concentrate, *Bulgur* had higher amount of carbohydrate (82.65%) and relatively high in fat. Wang &Murphy (1994) reported that protein, ash, fat and carbohydrate contents of soybean concentrate were 60%, 5%, 1% and 6% ,respectively.

The results in Table (1) show that *Bulgur* contained high values of the following types of dietary fibers; neutral detergent fiber (NDA) 31.33%, acid detergent fiber (ADF) 1.98%, hemicellulose (29.35%), cellulose (1.48%), and lignin (0.5%). These types of fibers are important elements in the human diet. They represent the unhydrolyzed plant storage and cell wall polysaccharides by human digestive enzymes (Bethesda, 1986). Moreover Yang *et al*. (2014) found that the total dietary fiber of *Bulgur* was 26%. This amount lies in the range of recommended amount by the American Diabetes Association in 1994 (25-35 g of dietary fiber per day) for persons suffering from diabetes and also in that suggested by the Cancer Institute in 1986 (20-35g /day) in US diet (National Cancer Institute, 1984).It has an immense effect on disease prevention and health maintenance, in addition to certain dietary fiber could act as a hypocholestrolemic factor (Wells & Ershoff, 1961).

Some function properties of *Bulgur* flour were determined and the results in Table (1) show that *Bulgur* had water absorption of 173.74%, fat absorption 175.87% and 39.0 ml oil /100g as emulsion stability. Determination of such properties helps in estimating the proper added quantity of dry *Bulgur* flour after hydration to replace minced meat in preparing some types of meat products. Generally, the functional properties of *Bulgur* flour are closely related to its content, compassion, presence, absence and availability of both hydrophilic and hydrophobic groups in its molecules. Also, its content of fat and fibers play a role in reducing and increasing such properties. Fat constrains water absorption while fibers encourage the holding capacity of water especially the soluble ones such as pectin and gums than cellulose fibers (Borderias *et al*., 2005). Notwithstanding, the capacity of fiber

Table 1: Proximate chemical composition, dietary fibers and some functional properties of *Bulgur* flour

Determination	Value*
1- Proximate composition (on dry weight basis%)	
Moisture	11.59± 0.010
Crude fat	1.80±0.05
Crude protein	13.74±0.09
Ash	1.81±0.05
Carbohydrate**	82.65±0.01
2- Dietary fibers (%)	
Neutral detergent fiber (NDF)	31.33±0.05
Acid detergent fiber (ADF)	1.98±0.04
Hemicellulose	29.35±0.04
Cellulose	1.48±0.03
Lignin	0.5±0.01
3- Functional properties	
Water absorption (%)	173.74±0.020
Fat absorption (%)	175.87±0.019
Emulsion stability (ml oil / 100g)	39.0±0.15

* Mean± S.D.   **Calculated by difference.

to bind fat depends more on its porosity than on its molecular affinity.

Isoflavone and antioxidant activity of *Bulgur* flour and soybean concentrate: The results in Table (2) show the HPLC separation, identification and content of isoflavones of *Bulgur* flour and soybean concentrate. The following isoflavones compounds were identified in *Bulgur* and soybean; genistein, biochanin A, diadzien and formononetin. The results indicated that soybean concentrate had high level of the different isoflavones comparing with *Bulgur* flour. Genistein, biochanin A, daidzien and formononetin in soybean concentrate were found to be more than 13, 6, 12 and 8 times than that in *Bulgur* flour. Wang & Murphy (1994) stated that isoflavones are one class of phytochemical compounds and found in large amounts in soybean. The major isoflavones in soybean are isoflavone aglycons, genistein and diadzein. Hendrich *et al.* (1994) reported that soybean is one of rich sources of isoflavones. Recently Kumar *et al.* (2013) mentioned that there is an argument about use of soybean in food production due to the similarity between chemical composition of its isoflavones and esterogen hormone.

Percentage of DPPH radical scavenging activity in both *Bulgur* flour and soybean concentrate was 28.84% and 54.42%, respectively as shown in Table (2) .This means that this characteristic in *Bulgur* was nearly half of soybean concentrate .This may be attributed to conditions of preparing *Bulgur* which include severe heat treatment during gelatinization   and drying of durum wheat comparing with these followed during soybean concentrate production including, solvent extraction and drying. Pratte & Birac (1979) found that soybeans, defatted soy flour, soy protein concentrates and soy isolate have appreciable antioxidant activity as detected by the rate of β-carotene bleaching in lipid –aqueous system. This was due to their content of phenolic compounds. According to Giambanelli *et al.* (2018), the antioxidant activity differed among durum wheat species. The whole einkon wheat had the highest antioxidant activity and the American whole emmer wheat had the lowest activity. According to Kumar *et al.* (2013) the antioxidant activity is mainly due to the phenolic compounds in the wheat grains such as flavonoids and the high reactivity of their hydroxyl groups. Acidity and processing such as boiling and cooking may cause up to 50% destruction in such compounds.

Beef burger characteristics and frozen storage stability

Proximate composition: The data in Table (3) show that the moisture, protein, fat, ash and carbohydrate contents ranged from 66.37 to 72.72, 59.13 to 70.86, 21.48 to 24.34, 3.78 to 4.81 and 0. 17 to 15.48%, respectively for the different types of the prepared beef burger investigated here. The highest significant ($P<0.05$) value of moisture, protein, fat, and ash contents were found in 100% meat beef burger and the lowest in the product containing 20% *Bulgur* flour. No significance ($P<0.05$)

Table 2: Isoflavones components and percentage of DPPH radical scavenging activity of *Bulgur* flour and soybean concentrate

Material	Isoflavones compounds as mg/g				DPPH radical scavenging activity (%)
	Genistein	Biochanin A	Daidzein	Formononetin	
Bulgur	0.0044	2.3735	1.1556	0.05408	28.84
Soybean	0.0554	13.4539	13.8053	0.4100	54.42

Table 3: Proximate chemical composition of beef burger samples (On dry weight basis).

Component (%)	Type of beef burger				
	Meat 100%	**Soybean concentrate 20%**	***Bulgur* flour**		
			10%	**15%**	**20%**
Moisture	72.72[a]	67.72[b]	68.83[b]	67.84[b]	66.37[c]
Crude protein	70.86[a]	66.34[b]	65.01[b]	62.09[c]	59.13[d]
Crude fat	24.34[a]	21.48[b]	22.98[ab]	22.30[ab]	21.61[b]
Ash	4.63[ab]	4.81[a]	4.06[ab]	3.92[b]	3.78[b]
Carbohydrate*	0.17[d]	7.37[c]	7.95[c]	11.69[b]	15.48[a]

*Carbohydrate was calculated by differences.
Means in a row with the same letter are insignificantly different at *P* < 0.05 level.

variations were traced in proximate composition between beef burgers containing 20% soybean concentrate and that with 10% *Bulgur* flour .Generally, replacing meat with *Bulgur* flour and soybean concentrate resulted in increase in carbohydrate content and reduction in other proximate composition components. Such changes were more noticeable with elevating the replacing meat level. This difference between soybean and *Bulgur* burger resulted from the difference in their chemical composition and the ratio of their incorporation.

Small differences were observed in the calculation of caloric value (kcal. 100g^{-1}) of these beef burger products. The burger (100% meat) was the highest in this value (503.18) followed by those contained 10% *Bulgur* (498.66), 15% *Bulgur* (495.82), 20% *Bulgur* (492.93) and lastly that having 20% soybean concentrate (488.16) .

Cooking loss, shrinkage and water holding capacity (WHC):- The data in Table (4) show significant (*P*<0.05) increase in cooking loss, shrinkage, and reduction in WHC of the prepared beef

Table 4: Cooking loss, shrinkage and water holding capacity (WHC) of beef burger samples after storage at -20°C for 3 months.

Property	Type of burger	Storage time at-20°C (month)		**Mean**
		0	**3**	
Cooking loss	100% Meat	22.96	29.90	26.43[a]
	20% Soybean concentrate	13.57	17.01	15.29[c]
	10% *Bulgur*	20.18	24.42	22.32[b]
	15% *Bulgur*	19.36	24.20	21.78[b]
	20% *Bulgur*	12.69	15.95	14.32[c]
Mean		17.75[b]	22.29[a]	
Shrinkage	100% Meat	40.82	45.75	43.28[a]
	20% Soybean concentrate	22.83	26.49	24.66[c]
	10% *Bulgur*	23.43	31.79	27.61[b]
	15% *Bulgur*	21.80	29.10	25.45[c]
	20% *Bulgur*	18.60	28.74	23.66[c]
Mean		25.49[b]	32.37[a]	
Water holding capacity (WHC %)	100% Meat	62.22	60.19	61.20[b]
	20% Soybean concentrate	67.70	65.82	66.76[a]
	10% *Bulgur*	68.62	64.47	66.54[a]
	15% *Bulgur*	68.34	64.57	66.45[a]
	20% *Bulgur*	68.12	64.71	66.41[a]
Mean		67.00[a]	63.95[b]	

Means with the same letter are insignificantly different at *P* < 0.05 level.

burger types after 3 months of storage at -20°C. Such changes were significantly (*P*<0.05) more in burger (contained 100% meat) and reduced with rise of plant meat replacer in the prepared beef burger types. No significant changes were observed between the two products containing 20% soybean concentrate and 20% *Bulgur* flour.

McDonagh *et al*. (2005) reported that normal fat beef burger had lower WHC and higher cooking loss. Shrinkage and cooking loss were found to increase during frozen storage due to muscle protein denaturation and release of water and fat. These are the main causes behind the reducing in piece diameter (shrinkage), WHC and cooking loss of burger. Plant meat substitutes reduced such losses by increasing binding forces between water and oil in this product. The reduction in cooking loss, WHC and shrinkage was observed with increase the proportion of plant meat replacer to 20%.Carbohydrate, especially fibers in *Bulgur*, protein and ash in soybean concentrate may be responsible for that (Hayes *et al*., 2009). Pintado *et al*. (2016) showed that cooking loss, shrinkage and WHC loss were lower in beef burger substituted with soybean flour, concentrate and isolate than the product free from such substitutes.

Colour, texture, pH and Thiobarbituric acid (TBA): The results in Table (5) show the changes in colour, texture, pH and TBA in beef burger samples at zero time and after 3 months of storage at -20°C.

Colour parameter: Substitution of meat by either *Bulgur* flour and /or soybean concentrate in preparing beef burger caused reduction in its fat and protein content (Table 3) and sequentially affected the colour parameters. Both lightness (L*) and yellowness (b*) increased while the redness (a*) of beef burger samples decreased due to plant substitution process. Relatively, changes in the aforementioned colour parameters were observed either for the type of plant substitutes, *Bulgur* and soybean concentrate, or for the ratio of meat replacer, in case of *Bulgur*. Storage at -20°C for 3 months caused an increase in lightness and relatively changes in redness (a*) and yellowness (b*) of beef burger samples containing plant replacer either *Bulgur* or soybean. Such changes were observed in burger containing 100% meat. Generally, frozen storage had little effect on the colour parameter of beef burger (Pintado *et al*. 2016).

Texture profile analysis : Texture properties were affected by substitution of meat with plant materials, *Bulgur* flour and soybean concentrate along with the frozen storage at -20°C for 3 months. As shown from Table (3), the burger (100% meat), rich in protein and fat than other products. This type of burgers, had the highest hardness before and after frozen storage (Table 5), comparing with the other burger products. This means that reduction in both fat and protein reduced the product hardness. This was confirmed when comparing the hardness of burger containing soybean concentrate with that containing *Bulgur* flour. The former contains high protein, fat and ash than *Bulgur* products. Fiber or carbohydrate in *Bulgur* may play a role in improving texture properties by increasing the emulsification stability and gelling properties of the *Bulgur* protein. Before frozen storage, the burger (100% meat), had higher cohesiveness than other's burger products containing plant meat substitutes. After 3 months of storage at -20°C, the carbohydrate or fibers in *Bulgur* burgers may be responsible for the increasing of their cohesiveness and chewiness.

The pH and TBA changes:- The results in Table (5) show that pH values ,ranged between 5.8 to 6.17 and 6.02 to 6.07 in different beef burger types before and after frozen storage for 3 months. These values were within the normal limits for the products of this kind. This means that the pH values of these products did not influence neither by type nor by the ratio of added plant meat replacer as well as frozen storage.

The TBA values of the different burger products before and after storage for 3 months at -20°C ranged from 0.008 to 0.018 mg malonaldehyde per1 kg sample. These values were below the level of incipient rancidity (≥ 1) (Ockerman, 1976). The low TBA values of different types of beef burger samples reported here, either free or containing *Bulgur* flour and soybean concentrate, before and after frozen storage, suggest that these products are viable in terms of oxidative stability.

Microbiological quality :- The Total Viable Count (TVC), Enterobacteriaceae, *E.coli*, *Coliform* and *Staphylococcus aureus* were determined in beef burger products during storage at -20°C for 3 months. The results in Table (6) show that TVC ranged from 3.5 x10³ to 6.0 x 10⁴ CFU/g in different burger products before and after frozen storage. Freezing temperature caused decreasing in the count of TVC in all samples. Generally this

Table 5 : Colour, texture, pH and TBA of beef burger samples after storage at -20ºC for 3 months

Type of burger	100% Meat		20% soybean concentrate		Bulgur					
					10%		15%		20%	
Storage time at –20ºC(month)	0	3	0	3	0	3	0	3	0	3
Property										
Colour units										
Lightness (L*)	39.88	45.38	43.66	47.71	43.92	47.90	44.02	49.35	44.78	50.80
Redness (a*)	6.46	4.39	5.73	5.42	5.15	4.14	5.34	4.16	5.96	4.33
Yellowness (b*)	14.93	14.39	16.39	14.66	16.89	14.96	16.84	14.14	16.88	14.40
Texture profile analysis (TPA)										
Hardness (g)	300	1465	294	869	283	893	262	905	252	921
Cohesiveness	0.66	0.00	0.59	0.58	0.50	0.54	0.53	0.61	0.56	0.64
Chewiness (mJ)	7.50	0.00	6.00	15.10	4.60	24.00	10.40	25.30	6.00	24.40
pH	5.80	6.05	6.17	6.07	5.89	6.03	5.86	6.03	5.87	6.02
TBA (mg malonaldehyde/Kg sample)	0.008	0.018	0.009	0.011	0.009	0.015	0.009	0.014	0.009	0.013

Table 6 : Microbiological quality of beef burger samples during frozen storage at -20ºC for 3 months

Bacterial count (CFU/g)	Type of burger	Storage time at -20°C(month)			
		0	1	2	3
TVC	100% Meat	6.0×10^4	3.9×10^3	4.2×10^3	3.8×10^3
	20% Soybean concentrate	5.3×10^4	4.7×10^3	3.8×10^3	4.1×10^3
	10% Bulgur	3.1×10^4	8.0×10^3	3.5×10^3	3.7×10^3
	15% Bulgur	3.5×10^4	5.1×10^3	4.6×10^3	3.5×10^3
	20% Bulgur	4.5×10^4	7.4×10^3	4.6×10^3	6.7×10^3
Enterobacteriaceae	100% Meat	5.1×10^3	2.0×10^2	N.D	N.D
	20% Soybean concentrate	7.2×10^2	4.0×10^2	N.D	N.D
	10% Bulgur	2.0×10^3	2.5×10^2	N.D	N.D
	15% Bulgur	2.4×10^3	2.0×10^2	N.D	N.D
	20% Bulgur	3.0×10^3	2.1×10^2	N.D	N.D

E. coli, coliform and *Staphylococcus aureus* were not detected in all samples.

N.D: Not detected

range of TVC population did not exceed the critical limit of 5×10^6 referred by the relevant 95/2 EC and 2073/2005 EC regulations for minced meat production, placing on market and meat preparation (EC, 1995 and EC 2005).

Enterobacteriaceae bacteria were only detected in very low load (7.2×10^2 to 5.1×10^3 CFU/g) in different burger samples after preparation then not detected after one month of storage at -20°C.This may be due to the effect of freezing temperature. *E.coli*, *Coliform* and *Staphylococcus aureus* were not detected in all samples either before or after 3 months of frozen storage .

Sensory evaluation:- The data in Table (7) show that all roasted beef burger samples containing *Bulgur* flour (10-20%) and /or soybean concentrate were judged as acceptable by the panelists. The beef burger product containing 10% *Bulgur* flour had significantly (*P<0.05*) the highest scores given by panalists for colour, taste, texture, odour and overall acceptability comparing with the other burger products. It was followed by burger containing 15% *Bulgur* flour, 20% *Bulgur* flour, 20% soybean concentrate and 100% meat product. This means that the reduction in fat to ~ 20% and protein to ~ 63% and an increase in fiber to 8-12 % in

Table 7 : Sensory evaluation of roasted beef burger samples during frozen storage at -20oC for 3 months

Property	Type of burger	Storage time at -20oC(month)				Mean
		0	1	2	3	
1- Colour	100% Meat	7.9	7.8	7.5	6.0	7.30[bc]
	20% Soybean concentrate	8.1	7.2	7.9	7.0	7.55[ab]
	10% *Bulgur*	8.0	7.9	8.2	8.0	8.02[a]
	15% *Bulgur*	8.3	7.9	8.1	7.0	7.82[ab]
	20% *Bulger*	7.6	7.6	6.9	5.6	6.94[c]
Mean		**7.98[a]**	**7.68[a]**	**7.72[a]**	**6.72[b]**	
2- Taste	100% Meat	7.8	7.0	7.2	5.6	6.91[b]
	20% Soybean concentrate	7.6	6.3	6.8	6.0	6.67[b]
	10% *Bulgur*	7.8	8.1	6.7	7.0	7.40[a]
	15% *Bulgur*	8.6	7.6	7.4	7.0	7.65[a]
	20% *Bulgur*	7.4	6.6	6.4	6.6	6.76[b]
Mean		**7.84[a]**	**7.12[b]**	**6.90[b]**	**6.44[c]**	
3- Odour	100% Meat	8.0	7.5	7.7	6.0	7.31[ab]
	20% Soybean concentrate	7.5	7.0	7.1	5.0	6.90[b]
	10% *Bulgur*	7.9	8.0	7.8	7.3	7.75[a]
	15% *Bulgur*	8.3	7.9	7.8	6.6	7.66[a]
	20% *Bulgur*	7.6	7.5	7.1	5.6	6.98[b]
Mean		**7.86[a]**	**7.58[a]**	**7.52[a]**	**6.10[b]**	
4- Texture	100% Meat	7.0	6.7	6.2	5.6	6.39[c]
	20% Soybean concentrate	7.8	7.3	7.5	5.6	7.06[b]
	10% *Bulgur*	7.9	8.1	7.8	7.0	7.70[a]
	15% *Bulgur*	8.3	7.6	8.2	7.0	7.75[a]
	20% *Bulgur*	7.9	7.6	7.1	6.0	7.75[a]
Mean		**7.78[a]**	**7.46[a]**	**7.34[a]**	**6.24[b]**	
5- Overall acceptability	100% Meat	7.2	7.1	6.3	5.8	6.60[b]
	20% Soybean concentrate	7.5	6.8	7.1	6.6	6.91[b]
	10% *Bulgur*	8.5	8.0	7.8	6.3	7.55[a]
	15% *Bulgur*	8.1	7.7	8.0	6.3	7.65[a]
	20% *Bulgur*	7.8	7.4	7.0	5.6	6.98[b]
Mean		**7.83[a]**	**7.33[b]**	**7.24[b]**	**6.12[c]**	

Means with the same letter are insignificantly different at $P < 0.05$ level.

burger formulation can produce acceptable product. Also, the panelists observed changes in colour, texture and odour of different beef burgers after 3 months of frozen storage at –20°C.

CONCLUSION

Replacing soybean concentrate with *Bulgur* flour did not affect quality and storage stability of beef burger. No differences were detected between their sensory evaluation, pH, TBA and microbiological quality before and after frozen storage at -20oC for 3 months.

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تأثير إحلال مركز فول الصويا بدقيق البرغل علي جودة والثبات التخزيني لبرجر اللحم البقري

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تم دراسة إحلال مركز فول الصويا بدقيق البرغل فى صناعة برجر اللحم البقري وأوضحت النتائج أن البرغل يحتوي على كربوهيدرات (٨٢,٦٥٪) وبروتين (١٣,٤٧٪) ورطوبة (١١,٥٩٪) و دهن (١,٨٪) ومحتوى الألياف الغذائية ٣١,٣٣ ٪ (NDF) و١,٩٨٪ (ADF). وكانت امتصاصية الماء وامتصاصية الدهن وثبات المستحلب ١٧٣,٧٤٪ و ١٧٥,٨٧٪ و ٣٩ مل / ١٠٠ جم زيت على الترتيب وأظهرت النتائج ان مركز فول الصويا يحتوي علي مستوى أعلي عن البرغل في محتوي Genistein, biochanin A, formononetin, daidzien بحوالي ١٣ضعفاً و٦ أضعاف و١٢ ضعفاً و٨ أضعاف وتم تقدير نشاط مضادات الأكسدة لكل من االبرغل ومركز فول الصويا فكانت ٢٨,٨٤٪ و٥٤,٤٢٪ على الترتيب. تم اعداد خمس عينات من البرجر هي : برجر خالٍ من الاضافات (كنترول) وبرجر يحتوي على دقيق البرغل بنسب ١٠و١٥و٢٠٪ وبرجر يحتوي علي مركز فول الصويا بنسبة٢٠٪ ، وقد أوضحت نتائج التركيب الكيماوي لعينات البرجر ان كلا من الرطوبة والبروتين والدهون والرماد والكربوهيدرات تراوحت من٦٦,٣٧الى٧٢,٧٢ ، ٥٩,١٣ الى ٧٠,٨٦، ٢١,٤٨ الى ٢٤,٣٤ ، ٣,٧٨ الى ٤,٨١ ، و١٧، ١٠الى ١٥,٤٨ ٪ على الترتيب وكان هناك ارتفاع واضح في كل من فاقد الطهي والانكماش بينما حدث انخفاض في قوة مسك الماء وذلك بعد ٣ شهور من التخزين علي –٢٠°م. و تراوحت قيم حمض الثيوباربيوتريك قبل وبعد التخزين لمدة ٣ شهورعلي –٢٠°م من ٠,٠٠٨الى ٠,٠١٨مجم مالون الدهيد / ١كجم عينة. وأظهرت نتائج الميكروبيولوجي ان كل العينات مقبولة من حيث العد الكلي المتاح حيث تراوح العد بين ٣,٥× ١٠³ و٦×١٠⁴ (CFU/g) سواءً قبل أو بعد التخزين لمدة ٣ شهورعلي –٢٠°م، أما الـ Enterbacteriaceae فقد تراوحت فى العينات قبل التخزين من ٧,٢×١٠² إلى ٥,١ × ١٠³ (CFU/g) واختفت بعد الشهر الأول من التخزين وكانت العينات خالية تماما من الـ E.coli والكوليفورم و Staphylococcus aureus سواءً قبل أو بعد التخزين ٣ شهورعلي –٢٠°م.وكانت العينات كلها مقبولة وفقا للاختبارات الحسية وكانت أفضل النتائج لعينات البرجر المضاف لها برغل بنسبتى ١٠٪ ، ١٥٪.