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CHEMICAL, PHYSICAL, MICROBIOLOGICAL AND ORGANOLEPTIC PROPERTIES OF LABORATORY-MADE BEEF MEAT PATTIES AS AFFECTED BY WHEAT GERM FLOUR

BY

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ABSTRACT

Ground beef patties formulated with wheat germ flour (WGF) 5%, 10% and 15% (as rehydrated 1:4 with water) was laboratory prepared. Physical, chemical and microbiological properties of the final products were determined. The obtained results showed that moisture and crude protein contents of fresh laboratory-made beef meat patties significantly decreased as the substitution level with dehydrated wheat germ fouler (DWGF) increased. On the other hand, ash and total carbohydrates contents markedly augment. Most of essential amino acids of fresh laboratory-made beef meat patties were gradually increased as the substitution level increased with exception of threonine and lysine. The first limiting amino acid was valine for control sample; whereas threonine was the limiting amino acid for beef patties contained DWGF. Physical and .organoleptic properties of fresh laboratory-made beef meat patties containing DWGF were not significantly different even at 10% substation level. Microbiological properties of fresh laboratory-made beef meat patties gradually decreased as the substitution level of DWGF increase. Thus, the lowest bacterial load was detected in beef patties contained 15% DWGF

Key words: ground beef patties, wheat germ flour, chemical and physical characteristics, Amino acids.

INTRODUCTION

Beef patties were popular and easy handling processed meat that ready to eat. At the past few decades, several investigations were studied the possibility to supplement comminuted meat products (CMP) with plant proteins to increase yield, improve stability, and modify textural properties (Martinez 1979, Lin & Zayas, 1987; Comer & Wojates, 1988 and Huang, *et al.*, 1999). In addition plant protein is effective not only to decrease the coast of the commodity but also lower fat and cholesterol content of the final products (Drake, *et al.*, 1975 and Morin *et al.*, 2002). The replacement of beef meat with plant proteins such as texturized soy bean, defatted wheat germ protein, corn protein, sorghum flour or lupine protein isolate in the ground meat led to increase the water retention in the final products (Rocha-Gara & Zayas, 1995; Huang *et al.*, 1999 and Khalifa, 2005). However the use of high levels of plant proteins in processed meat products may cause problems in color, and flavor (Mittal & Usborne, 1985 and Feng *et al.*, 2003).

This work was carried out to evaluate physical, chemical and microbiological properties of fresh laboratory-made beef meat patties contained wheat germ flour at different levels (5%, 10%, and 15%).

MATERIALS AND METHODS

Materials

Fresh wheat germ (FWG) was a gift from Middle and West Delta Milling Company at Tanta. Fresh beef meat (FBM) and fat (Tallow) were obtained from the local market of Alexandria City, Egypt. Other chemicals were obtained from EL-Nasr Company for pharmaceutical Chemicals, Egypt. Folin Dye Reagent and pyrogallol were purchased from Fluka Chemical (AG-CH9471). Thiobarbituric acid (TBA) was obtained from Sigma Chemical Co. (USA).

Methods

Preparation of laboratory-made beef meat patties

Fresh wheat germ (FWG) was subjected to steam in an autoclave for 10 min at 1.1 kg/cm^2 in order to inhibit all enzymes, especially, lipooxygenases. Then it was dried using an electric oven at 105°C, milled by a laboratory milling (Braun multi - Quick system 100 2k 100, Germany) to pass through 45m sieve to form dehydrated wheat germ flour (DWGF). The rehydrated wheat germ flour slurry (RWGFS) was prepared by rehydrating the DWGF with water at a ratio of 1:4 (W/V). The slurry was heated to 65°C on a magnetic hot plate with continuous stirring to gelatinize starch and increase water binding, and then held in a glass beaker for 90 min at room temperature to ensure complete hydration of DWGF (Huang *et al.*, 1999). The lean portion of FBM was trimmed. Both lean beef meat (90/10) and fat (95/5) were ground separately through a meat mincer (Braun multi - Quick system 100 2k 100, Germany). The Person Squire Calculation was used to determine the amounts of lean and fat portions needed to formulate patties of ground beef with 20% fat content. Ground fresh lean beef and fat were mixed again in a blender followed by cold water to achieve a uniform distribution of lean meat in the mixture. Ground beef (20% fat) was divided into 12 batches for three replications of each four treatments (control sample and beef meat patties contained 5, 10 and 15% DWGS) (Table, 1) and formed into about 113 g patties using a Hollymatic patty marker (Jet-Flow Super,; Park Forest, IL). Formed patties were placed between sheets of waxed paper and freezed at -20°C, packaged in polyethylene sheets and stored at -18°C until further analysis.

Table (1): Formulation of laboratory-made beef meat patties (20% fat) containing dehydrated wheat germ flour (DWGF)

Ingredient (g)	Laboratory-made beef meat patties			
	Control	Dehydrated wheat germ fouler		
		5%	10%	15%
Ground beef meat (80/20)	98	93	88	83
Wheat germ flour	–	5	10	15
Salt	2	2	2	2
Total (g)	100	100	100	100

Physical analysis

Water holding capacity (WHC) was measured according to the method of Huang *et al* (1999). Cooking yield was determined according to the method of Feng *et al.,* (2003). Water and Fat retentions were determined according Huang *et al.,* (1999) following the equations:

$$\%\ \text{Water retention} = \frac{\text{cooked patty weight} \times \%\ \text{moisture of cooked patty} \times 100}{\text{raw patty weight} \times \%\ \text{moisture of cooked patty}}$$

$$\%\ \text{Fat retention} = \frac{\text{cooked weight} \times \%\ \text{fat in cooked patty} \times 100}{\text{Raw weight} \times \%\ \text{fat in raw patty}}$$

Chemical analysis

Fresh beef meat patties were homogenized in food processor prior to sampling for proximate analysis. Moisture, ether extract, crude protein using micro-kjeldahl method (NX6.25), ash content as well as acid value, and peroxide value of fresh beef meat patties were determined according to the methods described in AOAC (1995). Amino acids composition was performed using Amino Acid Analyzer Instrument (Beakmann, Model 119 CL) at the Central Laboratory, Faculty of Agriculture, Alexandria University following sample digestion according to the method of Block *et al.,* (1958). The pH value of samples was measured using the method described by Bloukas *et al* (1997). Total volatile basic nitrogen (TVBN), trimethylamine (TMA) and Thiobarbituric acid (TBA) of samples were determined according to the method described by Pearson's (1981).

Microbiological Analysis

Total bacterial count, pyschropilic bacterial count and moulds & yeast were formed according to Difco manual (1984). Total Coliform count was detected on MacConkey broth media according to the method of Oxoide (1992). The results were recorded as most probable of coliform according to the index table tabulated in the manuals of quality control.

Sensory analysis

Sensory evaluation of cooked laboratory-made beef meat patties was carried out following standard sensory evaluation procedure (Meilgaard *et al.,* 1999). Twenty panelists were chosen from post graduate students and staff members from Food Science and Technology Department, Faculty of Agriculture, University of Tanta. The Panelists were asked to evaluate color, cereal or meat flavor, Juiciness and overall acceptability on a 1 to 15 hedonic scale A score of 1 being dislike extremely and 15 being like extremely.

Statistical Analysis

Physical, chemical analysis of beef patties were performed in triplicate. The obtained results were analyzed statistically as a completely randomized block design using SAS procedures (SAS Institute, Inc., 1988). Means separation were performed using Duncan's Multiple Range Test method (Ott, 1988).

RESULTS AND DISCUSSION

Data of Table (2) show that crude protein (N × 6.25) and ether extract contents of FBM were significantly ($p<0.05$) higher than that of fresh wheat germ flour (DWGF). Contrary to ash and total carbohydrates of FBM were significantly lower than those of DWGF. In the meantime, no significant differences were found between pH value of FBM and DWGF. As microbial aspect it is obvious that FBM involved different microorganisms while DWGF was free from such microorganisms. This may be related to the heat of autoclaving and dehydration temperatures which were performed during preparation of DWGF from the fresh wheat germ. These results are in agreement with Khalifa (2005).

Table (2): Proximate analysis (g/100g on dry weight basis), pH value and microbial properties of fresh beef meat (FBM) and dehydrated wheat germ flour (DWGF)

Analysis	Fresh beef meat (FBM)	Dehydrated wheat germ flour (DWGF)
Approximate analysis		
Crude Protein (%)	76.1 ± 1.9*	31.5 ± 0.30
Ether extract (%)	21.8 ± 1.1*	18.7 ± 1.85
Ash (%)	1.6 ± 0.5*	10.9 ± 0.41
Total carbohydrates (%)	0.5 ± 0.1*	38.9 ± 1.95
pH value	6.4 ± 0.1	5.7 ± 0.1
Microbial aspects		
Total plate count (cfu g^{-1})	5.1×10^5	ND
Moulds &Yeast (cfu g^{-1})	3.0×10^3	ND
Psychrophilic Bacteria (cfu g^{-1})	7.1×10^3	ND
Coliform Group (cfu g^{-1})	ND	ND

Total carbohydrates were calculated by difference

M± SD = means and standard deviation of three replicate

* Significantly different at 5% level ND = Not Detected

Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on approximate analysis of fresh laboratory-made beef meat patties:

Moisture and protein contents of fresh laboratory-made beef meat patties were decreased as the substitution level with DWGF increased (Table, 3). This may be related to FBM contains higher percentage of moisture and protein contents than that of DWGF beside of the hydrophilic property of plant proteins present in the DWGF (Rechardjo *et al.*, 1994). On the other hand, ash and total carbohydrates contents of fresh laboratory-made beef patties (control) were gradually raised as the substitution level with DWGF increased. The increment in both ash and total carbohydrates contents of substituted laboratory-made beef meat patties may be related to the property of DWGF where, it contains more carbohydrates and ash rather than that of FBM. Such these results were reported in the literature (Frances *et al.*, 1978; Ketton & Melton, 1978; Nofal, 1981; El-Akary, 1986; El–Wakeil *et al.*, 1994 and Khalifa, 2005).

Table (3): Proximate analysis (g/100g on dry weight basis) of fresh laboratory-made beef meat patties as affected by substitution FBM with DWGF at different levels (5, 10 and 15%)

Samples of beef patties	Moisture	Crude Protein	Ether Extract	Ash	* Total Carbohy-drates
Control	75.5 ± 2.4^{a}	74.9 ± 3.6^{a}	20.5 ± 1.6^{a}	2.7 ± 0.2^{c}	1.9 ± 0.7^{d}
5% DWGF	75.1 ± 11.7^{a}	74.1 ± 14.1^{a}	19.7 ± 3.8^{a}	2.9 ± 1.6^{c}	3.3 ± 1.7^{c}
10% DWGF	72.2 ± 11.7^{ab}	70.6 ± 14.1^{b}	18.8 ± 3.8^{ab}	3.3 ± 1.7^{b}	7.3 ± 1.8^{b}
15% DWGF	70.0 ± 11.7^{b}	68.2 ± 14.1^{c}	18.3 ± 3.8^{b}	4.7 ± 2.0^{a}	8.8 ± 1.7^{a}

*Total carbohydrate were calculated by difference
M±SD= Mean and standard deviation of three replicates
In a column, means having the same common superscript letter are not significantly different at 5% level.

Most of essential amino acids of fresh laboratory-made beef meat patties were gradually increased as the substitution level increased with exception of threonine and lysine (Table, 4). Thus, the raise the substitution level in the laboratory-made beef meat patties, the higher the essential amino acid content except both threonine and lysine. This may related to the lack of both amino acids in DWGF, as plant protein, comparing with beef meat.

The first limiting amino acid in fresh laboratory-made beef meat patties was valine (chemical score 0.60) for control (Table, 5). While threonine is the limiting amino acid for fresh laboratory-made beef meat patties contained DWGF at different levels (5, 10 and 15%).

Total volatile nitrogen (TVN), trimethylamine (TMA) of fresh laboratory-made beef meat patties significantly ($p<0.05$) diminished by rising the substitution level with DWGF. In contrast, pH value increased as the substitution level with DWGF augmented (Table, 6). The increment of pH value may be related to the specificity of plant protein and its alkaline ash (Susan *et al.*, 1992, Karen *et al.*, 1997, Huang *et al.*, 1999). While the decrement in TVN and TMA could be related to low amounts of volatile nitrogen of plant proteins (El-Akary, 1986 and Khalifa, 2005).

Acid value (AV), peroxide value (PV) and thiobarbituric acid value (TBA) of lipid extracted from fresh laboratory-made beef meat patties were not significantly ($p<0.05$) different at 5% DWGF substitution level than that of control one. However, these values were significantly decline at substitution level 10% and 15% with DWGF. These results are in agreement with Liu *et al.* (1991) and Khalifa (2005).

Table (4): Amino acid composition (g AA/100g protein) of fresh laboratory - made beef meat patties as affected by substitution FBM with DWGF at different levels (5, 10 and 15%)

Amino acid	Control	Dehydrated wheat germ flour (DWGF)		
		5%	10%	15%
Essential				
Leucine	5.91	6.10	6.16	6.74
Valine	**3.26**	4.24	4.57	5.67
Threonine	3.75	**2.81**	**2.34**	**2.01**
Methionine	4.40	4.71	4.82	4.88
Phenylalanine	5.11	5.55	5.57	5.20
Lysine	5.16	4.61	4.53	4.16
Non-Essential				
Aspartic	7.03	7.06	8.23	8.10
Serine	3.51	3.66	3.54	3.22
Glutamic	10.39	12.14	13.44	14.20
Proline	12.15	8.60	7.33	7.05
Glycine	4.42	4.47	4.93	5.00
Alanine	3.90	5.00	5.98	6.00
Cysteine	0.50	0.30	0.20	0.20
Isoleucine	3.81	4.31	4.51	4.49
Tyrosine	4.68	3.53	3.93	4.12
Histidine	5.09	4.02	3.88	3.75
Arginine	4.21	7.81	8.88	9.74
	87.28	**88.92**	**92.84**	**94.53**

Essential amino acid was classified according to Sanders and Amery (2003)

Table (5): Chemical score and limiting amino acid of fresh laboratory -made beef meat patties substituted FBM with DWGF at different levels (5%, 10% and 15%)

Essential amino acid	Control	Dehydrated wheat germ flour			FAO (1974)
		5%	10%	15 %	
Leucine	0.89	0.90	0.91	0.97	8.85
Valine	**0.60**	0.76	0.82	1.00	7.26
Threonine	0.98	**0.72**	**0.60**	**0.51**	5.07
Methionine	1.85	1.95	1.78	1.98	3.15
Phenylalanine	1.16	1.24	1.25	1.14	5.84
Lysine	1.06	0.93	0.92	0.82	6.45

Essential amino acid was classified according to Sanders and Amery (2003)

Table (6): Influence of substitution FBM with DWGF at different levels (5, 10 and 14%) on some chemical properties of fresh laboratory-made beef meat patties.

Samples	pH Value	TVN* $mg100g^{-1}$	TMA** $mg100g^{-1}$	AV◆ $mg100g^{-1}$	PV† $meqO_2\ kg^{-1}$)	TBA‡ $mg\ kg^{-1}$
Control	6.10 ± 0.1^{c}	51.0 ± 7.1^{a}	7.3 ± 1.3^{a}	3.1 ± 0.5^{a}	12.0 ± 1.1^{a}	1.1 ± 0.3^{a}
5%DWGF	6.25 ± 1.3^{b}	41.4 ± 5.4^{b}	6.1 ± 1.6^{b}	3.1 ± 0.6^{a}	11.8 ± 3.4^{a}	1.0 ± 0.3^{ab}
10%DWGF	6.47 ± 1.4^{b}	35.1 ± 5.2^{c}	4.9 ± 1.6^{c}	2.7 ± 0.7^{b}	10.5 ± 3.4^{b}	0.9 ± 0.1^{b}
15%DWGF	7.01 ± 1.3^{a}	30.7 ± 5.4^{d}	4.1 ± 1.6^{d}	2.2 ± 0.6^{b}	10.0 ± 4.3^{b}	0.9 ± 0.1^{b}

M±SD = means and standard deviation of three replicates AV◆= Acid Value
TVN* = Total volatile nitrogen TMA** = Trimethylamine
PV†= Peroxide value TBA‡= Thiobabuturiv value
In a column, means having the same superscript letters are not significantly different at 5% level

Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on physical characteristics of fresh laboratory-made beef meat patties

Water-holding capacity (WHC) of laboratory-made beef meat patties significantly ($p<0.05$) decreased due to the presence of DWGF in the formula of patties. Since, the higher the DWGF content in beef meat patties, the lower the WHC. On the other hand, cooking yield, water and fat retentions of fresh laboratory beef meat patties were significantly ($p<0.05$) increased as the substitution level of DWGF raise (Table, 7).

Table (7): Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on some physical properties of fresh laboratory-made beef meat patties.

Samples	WHC (%)	Water Retention (%)	Fat Retention (%)	Cooking Yield (%)
Control	61.6 ± 07.3^{c}	58.1 ± 5.3^{c}	45.3 ± 3.7^{c}	74.9 ± 06.1^{d}
5% DWGF	69.0 ± 16.1^{b}	61.8 ± 5.1^{b}	48.7 ± 3.7^{a}	77.6 ± 05.4^{c}
10% DWGF	72.6 ± 16.2^{ab}	63.0 ± 5.3^{a}	47.3 ± 3.5^{ab}	77.7 ± 17.2^{b}
15% DWGF	73.5 ± 16.6^{a}	65.1 ± 5.3^{a}	46.7 ± 3.7^{b}	79.4 ± 17.5^{a}

M±SD = means and standard deviation of three
In a column, means having the same superscript letters are not significantly different at 5% level

The decrement in WHC of laboratory-made beef meat patties may be attributed to the protein denaturation and/or aggregation, which markedly affect on its chemical bonds especially the functional groups responsible for water binding properties (El-Akary, 1986). Where, the increment in cooking yield, water and fat retention could be attributed to the capability of plant protein to bind higher moisture and fat content rather than beef protein. In this respect, Rocha-Garza and Zayas (1995) indicated that the wheat germ protein flour acts as a binder for moisture and fat in finely ground meat. These results are in agreement with those reported by Huang *et al.*, (1999) and Su *et al.*, (2000).

Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on sensory evaluation of fresh laboratory-made beef meat patties

Sensory evaluation of fresh laboratory-made frankfurter was markedly influenced by the presence of DWGF. Whereas color, firmness; and meat flavor panelist's scores of beef meat patties were significantly ($p < 0.05$) diminish by increase the substitution level of DWGF (Table, 8). On the other side, juiciness and cereal flavor of the final product was significantly ($p \leq 0.05$) progress as the substitution level of DWGF raised. The decrease in color score of prepared beef meat patties contained DWGF could be related to dilute the meat pigments and the ability of particle of plant protein to adsorb meat juice (Judge *et al* 1974). The improvement in firmness could be related to augment tissue destruction by higher pumping level and the high level of water in this treatment (Siegel *et al.*, 1979). These results perfectly agree with the literature (Bloukas *et al.*, 1997 and Lin & Mei, 2000; Morin *et al.*, 2002; and Khalifa, 2005).

Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on microbiological properties of fresh laboratory-made beef meat patties

Total bacterial count of fresh laboratory-made beef meat patties gradually decreased as the substitution level of DWGF in beef meat patties increase (Table, 9). The same trend was also observed for coliform, psychrophilic bacteria and molds & Yeast counts. Therefore, the lowest microbial count was detected in beef meat patties contained 15% DWGF. Such results were obtained by Khlifa (2005).

Table (8): Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on some sensory evaluation of fresh laboratory-made beef meat patties

Samples	Color	Firmness	Juiciness	Cereal flavor	Meat flavor
Control	12.2 ± 1.5^{b}	14.2 ± 0.6^{a}	11.8 ± 2.7^{c}	1.1 ± 0.7^{a}	14.5 ± 0.5^{a}
5% DWGF	12.4 ± 1.7^{b}	13.9 ± 0.6^{a}	12.1 ± 0.7^{b}	1.8 ± 0.7^{b}	13.7 ± 0.5^{a}
10% DWGF	13.3 ± 1.7^{a}	12.3 ± 0.8^{ab}	13.1 ± 0.7^{ab}	7.9 ± 0.9^{c}	8.0 ± 0.7^{b}
15% DWGF	$14. \pm 0.9^{a}$	9.3 ± 1.0^{c}	14.0 ± 0.6^{a}	12.3 ± 1.0^{d}	7.1 ± 0.8^{c}

M ±SD = mean ± standard deviation

In a column, means having the same superscript letters are not significantly different at 5% level

Table (9): Influence of substitution FBM with DWGF at different levels (5, 10 and 15%) on microbiological aspects of fresh laboratory-made beef meat patties (cfu g^{-1})

Samples	Total Count	Coliform Bacteria	Psychrophilic Bacteria	Molds & Yeast
Control	1.8×10^5	1.4×10^3	9.5×10^2	13.6×10^3
5% DWGF	1.5×10^5	4.5×10^2	8.3×10^2	9.5×10^2
10% DWGF	1.4×10^4	4.3×10^2	7.4×10^2	7.8×10^2
15% DWGF	1.1×10^4	1.1×10^2	4.0×10^2	5.6×10^2

In conclusion, wheat germ is the by-product of wheat milling and contains valuable components including high quality proteins, minerals, essential fatty acids, sterols and vitamins which prevent from coronary heart diseases. The addition of wheat germ flour to beef patties reduces the cholesterol content and improves physical properties, microbial aspects and sensory quality of the final product. Wheat germ flour can be added up to 5% (W/W) for the production of beef patties.

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تأثير اضافة دقيق جنين القمح علي الصفات الكيماوية والطبيعية والميكروبيولوجية والحسية العضوية لأقراص اللحم المحضر معمليا

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تم تحضير أقراص اللحم المحتوية علي دقيق جنين القمح المبلل (بنسبة ١:٤ جنين قمح :ماء) في المعمل. تم قدرت الصفات الكيماوية والطبيعية والميكروبيولوجية وكذلك الصفات الحسية العضوية لأقراص اللحمة الطازجة لمعرفة تأثير أستبدال دقيق جنين القمح بالنسب المختلفة (٥ و١٠ و١٥٪). وقد أوضحت النتائج المتحصل عليها أن نسبة الرطوبة والبروتين الخام لأقراص اللحمة تنخفض معنويا كلما زادت كمية الأستبدال بواسطة دقيق جنين القمح. بينما زادت نسبة كل من الرماد والكربوهيدرات زيادة معنوية بإرتفاع نسبة الأستبدال. كذلك وجد أن نسبة الأحماض الأمينية الضرورية في بروتينات أقراص اللحم المحضر معمليا تزداد بزيادة نسبة دقيق جنين القمح بأستثناء الاحماض الامينية الثريونين والليسين. وكان الحمض الأميني المحدد في أقراص اللحم البقري دون استبدال هوالفالين بينما كان الحمض الأميني ثريونين هوالحمض الأميني المحدد لأقراص اللحم المحتوية علي دقيق جنين القمح. أظهرت النتائج تحسن في الصفات الطبيعية والحسية العضوية لأقراص اللحم بزيادة نسبة دقيق جنين القمح المستبدل. لوجظ أن الحمل الميكروبي لأقراص اللحم المحضر معمليا ينخفض بزيادة نسبة دقيق القنح في الخلطة حيث كان أقل حمل ميكروبي موجودا بأقراص اللحم المحتوية علي ١٥٪ دقيق جنين القمح.