Research Article Effect of Lupine Flour on Baking Characteristics of Gluten Free Cookies

 ¹Sofyan Maghaydah, ²Selma Abdul-hussain, ³Radwan Ajo, ¹Yousef Tawalbeh and ¹Noor Elsahoryi ¹Department of Nutrition and Food Technology, Faculty of Agriculture, Jordan University of Science and Technology, P.O. Box 3030, Irbid 22110, Jordan
 ²Food Science and Biotechnology Department, Faculty of Agriculture, Baghdad University, Iraq ³Applide Science Department, Al-Huson University Collage, Al-Balqa Applied University, P.O. Box 50, Al-Huson 21510, Jordan

Abstract: Celiac Disease (CD) is an immune-mediated disease in genetically susceptible individuals caused by intolerance to gluten protein in some cereals, resulting in mucosal inflammation, which causes malabsorption. An effective treatment for CD is a gluten-free diet that excludes cereals containing gluten. One of the most desirable wheat products is the cookie, which is considered suitable for all ages due to its low manufacturing cost, convenience, long shelf life and good eating quality. Therefore, the production of local, high quality and affordable gluten-free cookies was the main objective of this study in which lupine flour was used as a main wheat flour alternative. Eight gluten-free cookie flour blends were prepared: 100% Lupine Flour (AF), 50% lupine flour and 50% corn starch (BF), 40% lupine flour and 60% corn starch (CF), 30% lupine flour and 70% corn starch (DF), 30% lupine flour, 40% rice flour and 30% corn starch (EF), 30% lupine flour, 40% corn flour and corn starch 30% (FF), 20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch (GF) and 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (HF), with equal amounts of hydrocolloids (1.5% xanthan and 1.5% carrageenan) which were used as a functional gluten alternative, as well as a control sample with Wheat flour (WC). The chemical composition, physical characteristics and sensory evaluation of all treated flour blends and cookies were determined. The results of the chemical analysis indicated that corn and wheat flour were significantly $(p \le 0.05)$ higher in moisture content, while lupine flour had significantly $(p \le 0.05)$ higher contents of lipid, protein, fiber and ash. Starch significantly ($p \le 0.05$) showed the highest carbohydrate content. The moisture of blend BC was significantly ($p \le 0.05$) higher than all blends and blend AC was significantly ($p \le 0.05$) higher in ash, protein, lipid and fiber content. The carbohydrate content of blend DC was significantly ($p \le 0.05$) higher than the other blends, in which blend AC significantly (p≤0.05) contained the lowest amount. Physical analysis revealed that the spread factor of blend GC was significantly ($p \le 0.05$) higher than the other blends, while blend CC significantly ($p \le 0.05$) had the lowest value. The results of color measurements (L*, a* and b*) using a Milolat colorimeter revealed that the L* parameter had the highest value in the control sample WC, while the a* parameter was highest in blend AC and b* was highest in blend EC. The results of the sensory evaluation, judged by panelists, indicated that blend BC received the highest score in overall impression, while the control sample WC received the overall highest score in flavor and crust color, blend GC in overall color and the control sample WC in overall texture, blend AC in crust color, crumb color, hardness and after taste. Finally, blends BC and GC had the best acceptability compared to the control sample.

Keywords: Celiac disease, corn flour, corn starch, gluten free cookies, lupine flour, rice flour

INTRODUCTION

Celiac Disease (CD) is an immune-mediated disease that occurs in genetically susceptible individuals and is caused by intolerance to ingestion of gluten, a protein found in wheat, barley, rye and perhaps oat (Rubio-Tapia and Murray, 2010). It is a chronic non-infectious disorder with an incidence of about one in every 300 births. CD is characterized by mucosal inflammation, thrive failure, weight loss, hypotonia, abdominal distention, steatorrhea and malabsorption caused by damaged small intestine villi (Beers and Berkow, 2004). An effective treatment for CD is a gluten-free diet that excludes cereals containing gluten (Beers and Berkow, 2004). Wheat is the main ingredient in many foods such as breads, breakfast cereals, breaded foods, crackers, pretzels, pastas and cookies (Hussain *et al.*, 2006). Therefore, avoiding wheat is probably the biggest challenge for people with CD. Cookies are baked products which are considered

Corresponding Author: Sofyan Maghaydah, Department of Nutrition and Food Technology, Faculty of Agriculture, Jordan University of Science and Technology, P.O. Box 3030, Irbid 22110, Jordan

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the most desirable products for all ages due to their low manufacturing cost, convenience, long shelf life and good eating quality. Moreover, cookies could be used as a vehicle to deliver essential nutrients to people. The production of high quality gluten-free cookies is a challenge considering the fact that gluten is responsible for the viscoelastic properties and formation of cohesive dough (Kulp and Ponte, 2000). Therefore, the production of gluten-free cookies requires choosing wheat flour alternatives that have similar functional properties as wheat flour and which may also be more nutritious (Hoseney, 1986). Based on preliminary work, lupine bean (Lupineus albus) can be used as a base in gluten-free cookies due to its unique physical and chemical properties compared with other wheat flour alternatives, rice and corn flour. The nutritional value of lupine is distinctive in comparison with other cereal flour as it contains high amounts of protein (36-52%), oil (5-20%) and fiber (30-40%) (Mohamed and Rayas-Duarte, 1995), as well as vitamins such as thiamin, riboflavin, niacin, folate and vitamin E (Erbas et al., 2005). Lupine is also rich in many nutritional compounds such as phytoestrogens, phytosterols and antioxidants. Therefore, frequent consumption of lupine could help protect against cancer, cardiovascular hypertension, diabetes mellitus disease, and osteoporosis. In addition, rice and corn were selected due to their functional properties during dough formation. Currently, the cereal products designed to meet the requirements of celiac disease patients, especially gluten-free cookies, are scant, limited, low quality and provide poor mouth-feel and flavor, in addition to their high cost in Jordan. Therefore, utilization of lupine flour to improve the quality of gluten-free cookies (possibly with enhanced nutrition) was the main objective of this study.

MATERIALS AND METHODS

Materials: Patent wheat flour was used to produce the cookies as the control. Corn flour, rice flour and corn starch were obtained from Modern Flour Mills and Macaroni Factories (Amman, Jordan). Sweet lupine beans (*Lupineus albus*) were purchased from the local market and were milled into flour using a Labconco mill (Laboratories Construction, Kansas City, MO) adjusted to give coarse particles. Hydrogenated vegetable shortening, lecithin, sugar, milk, sodium bicarbonate, salt and two commercial food-grade gum samples, xanthan and carrageenan were purchased from Guangdong Guanghua Chemical Factory, China.

Lupine bean milling: According to Al-Omari (2009) method, lupine beans were cleaned (discarding small, broken, moldy and damaged beans), then washed with distilled water and then sterilized with 0.3% sodium hypochlorite solution for 1 min, then rewashed again with distilled water, then soaked in distilled water for 20 h and finally dried in a ventilated oven (Vindon, England) at 55°C for 18 h. The dried beans were

ground using an electrical mill (Braun, Germany) to pass through a 60 mesh sieve (British standard screen). The milled flour was stored in air-tight polyethylene bags at 4°C until required.

Cookie flour: Eight gluten-free flour blends were used in this study to produce gluten-free cookies. That blends included: 100% lupine flour (AF) and its corresponding cookies indicated as (AC), 50% lupine flour and 50% corn starch (BF) and its resulting cookies indicated as (BC), 40% lupine flour and 60% corn starch (CF) and its corresponding cookies indicated as (CC), 30% lupine flour and 70% corn starch (DF) and its corresponding cookies indicated as (DC), 30% lupine flour, 40% rice flour and 30% corn starch (EF) and its corresponding cookies indicated as (EC), 30% lupine flour, 40% corn flour and 30% corn starch (FF) and its corresponding cookies indicated as (FC), 20% lupine flour, 30% corn flour and 20% rice flour and 30% corn starch (GF) and its corresponding cookies indicated as (GC). The control cookies were produced from Wheat flour (WC).

Cookie preparation: The basic gluten-free cookie recipe and preparation procedure were based on Obeidat et al. (2012). The basic ingredients used were gluten-free flour (250 g), sugar (100 g), shortening (90 g), milk (25 g), sodium bicarbonate (1.0 g), salt (2.5 g), lecithin (1.25 g), xanthan (3.75 g), carrageenan (3.75 g) and water as required. Sugar and shortening were mixed thoroughly in a bowl for 2 min at speed four using a mixer (Kenwood[®], Britain). The dry ingredients were weighed and mixed with water for 3 min at speed 3 to get cookie dough. The dough was rolled thinly on a sheeting board to a uniform thickness (6.0 mm) and cut out using a round steel cookie cutter (35.0 mm diameter). The cut-out cookie dough pieces were baked on greased pans at 180°C for 12-15 min in a baking oven (Rational[®], Germany) and then were allowed to cool at room temperature (25±2°C) for 8-10 min. The control cookies prepared using patent wheat flour extraction rate (45%) using the same formula without xanthan and carrageenan gums and without the addition of lupine flour. All cookies were stored in air-tight containers until evaluation.

Chemical analysis: The Standard Association of Official Analytical Chemistry methods, (AOAC, 1995) were adopted for estimating moisture, ash, crude fiber, protein and fat contents. Total carbohydrates were calculated as 100- (protein+fat+moisture+ash+fiber).

Physical analysis: Published methods (AACC, 2000) were used to evaluate the width, thickness and spread factor of the cookies. The Width (W) was measured by placing six cookies edge to edge to get an average width in millimeters. The Thickness (T) was measured

by stacking six cookies on top of one another, then restacking in a different order and measuring them to get the average in millimeters. The Spread Factor (SF) was determined by the following equation:

SF = W/T

Color measurement: The color of cookie samples were measured using a Minolat colorimeter CR-300 (Ramsey, N.J., U.S.A) and recorded using the L*a*b* color system. The L*a*b* color system consists of a luminance or Lightness component (L*) and two chromatic components: the (a*) component for green (-a) to red (+a) and the (b*) component from blue (-b) to yellow (+b). The colorimeter was calibrated using a standard white plate. The values of white the standard were L = 97.1, a = +0.13, b = +1.88. Color was measured at two positions on both sides. Triplicate samples were used for each blend and then the measurements were averaged.

Trained organoleptic evaluation: Ten panelists were trained by the researcher using standard product evaluation criteria developed by Hussain *et al.* (2006). Two training sessions were conducted in which the panelists were trained to evaluate the sensory attributes of the gluten-free cookies. The panelists used the orientation session to improve their reproducibility and accuracy. Randomly coded samples were served to the panelists individually. Panelists were supplied with drinking water for cleansing the palate between samples. The sensory evaluation was carried out on the overall impression, overall flavor, overall texture, overall color, crust color, crumb color, hardness and aftertaste of the sample. A hedonic scale test was used for overall impression, flavor, texture and crust color of

Table 1: Chemical composition of different flour types

the sample: 1 = dislike extremely to 9 = like extremely. A descriptive scale was used for all other parameters.

Statistical analysis: The data were analyzed using the Statistical Package for Social Sciences (SPSS, version 15.0, 2007, Chicago, IL). One-way Analysis of Variance (ANOVA) was performed to test differences between blends followed by mean separation using Duncan's analysis. Findings with a p-value of ≤ 0.05 were considered to be statically significant.

RESULTS AND DISCUSSION

Chemical analysis: The chemical composition of all types of flour (wheat, rice, corn and lupine) and corn starch is shown in Table 1. Wheat, rice and corn flour contained the highest moisture content, while the corn starch and lupine flour had the lowest. Lupine flour had the highest content of protein (33.3%), lipid (8.87%), fiber (10.75) and ash (1.82), but the lowest carbohydrate content (36.76%). Mean while, corn starch had the highest content of carbohydrates (89.47%) and the lowest protein content (0.02%).

The results of the chemical analysis of cookies are shown in Table 2. There was a significant difference among all blends with regards to moisture content. Moreover, blend BC contained the highest water level (3.55%), since it had the highest content of starch and 50% lupine, both of which have high water holding capacity due to the presence of polar amino acids and hydroxyl groups, respectively. Kohajdová *et al.* (2011) reported that lupine protein shows good water and fat binding capacity; this agrees with the chemical analysis results for the moisture and lipid contents of the

Tuble 1. Chemical composition of american hour types								
Flour type	Moisture %*	Ash %*	Protein %*	Lipid %*	Fiber %*	Carbohydrate %**		
Wheat	11.21±0.5 ^a	0.40±0.11°	9.70±0.14 ^b	1.16±0.04°	0.11±0.01°	77.42°		
Rice	10.93±0.1 ^a	0.31±0.06 ^c	$7.40\pm0.45^{\circ}$	0.97 ± 0.11^{cd}	$0.02{\pm}0.00^{d}$	80.37 ^b		
Corn	11.21±0.4 ^a	0.59±0.01 ^b	6.85±0.55°	1.80±0.22 ^b	0.29±0.01 ^b	79.26 ^b		
Lupine	8.50±0.4°	1.82±0.01 ^a	33.30±0.03ª	8.87 ± 0.16^{a}	10.75±0.07 ^a	36.76 ^d		
Corn starch	9.71±0.3 ^b	0.13 ± 0.20^{d}	$0.02{\pm}0.00^{d}$	0.66 ± 0.07^{d}	$0.01{\pm}0.00^{d}$	89.47 ^a		

*: Values are the average of two replicates \pm the standard deviation; **: Percent calculated by the difference; Different superscript letters in the same column indicate a significant (p \leq 0.05) difference according to Duncan's test

Table 2: Chemical	composition of	cookies p	produced from	n different flour	blends

ruble 2. Chemieur composition of cookies produced nom unicient nour biends							
Cookie blends	Moisture %*	Ash %*	Protein %*	Lipid %*	Fiber %*	Carbohydrate %*	
WC	0.98±0.70 ^g	1.30±0.01 ^d	7.36±0.19 ^e	18.42 ± 0.02^{d}	1.23 ± 0.04^{f}	70.71 ^a	
AC	$3.04{\pm}0.02^{b}$	3.01±0.14 ^a	18.90±0.06 ^a	23.65±0.37 ^a	6.45 ± 0.07^{a}	44.95 ^e	
BC	3.55±0.02 ^a	1.83±0.01°	16.32±0.71 ^b	20.60±0.04 ^b	1.89 ± 0.09^{d}	55.93 ^d	
CC	1.58 ± 0.05^{f}	2.02±0.01°	10.40 ± 0.14^{d}	20.29±0.29 ^b	$1.94{\pm}0.07^{d}$	63.77 ^c	
DC	1.93±0.02 ^e	$1.98\pm0.01^{\circ}$	10.04 ± 0.61^{d}	18.53 ± 0.22^{d}	1.39 ± 0.14^{f}	66.13 ^b	
EC	1.04±0.01 ^g	1.82±0.01°	12.34±0.10 ^{cd}	18.93 ± 0.14^{d}	1.70±0.05 ^e	64.17 ^c	
FC	2.22 ± 0.04^{d}	1.86±0.01°	11.77±0.20 ^{cd}	19.68±0.00°	1.30 ± 0.14^{f}	63.22 ^c	
GC	2.95±0.01 ^{bc}	2.53±0.38 ^b	13.02±0.11°	20.72 ± 0.98^{b}	2.92 ± 0.02^{b}	57.86 ^d	
HC	2.74±0.04°	1.76±0.01°	13.54±0.11°	18.37 ± 0.44^{d}	2.74±0.08°	60.86 ^c	

*: Values are the average of two replicates \pm the standard deviation; **: Percent calculated by the difference; Different superscript letters in the same column indicate a significant (p \leq 0.05) difference according to Duncan's test; WC: 100% wheat flour (control); AC: 100% Lupine Flour (LF); BC: 50% lupine flour and 50% corn starch (BF); CC: 40% lupine flour and 60% corn starch (CF); DC: 30% lupine flour and 70% corn starch (DF); EC: 30% lupine flour, 40% rice flour and 30% corn starch (EF); FC: 30% lupine flour, 40% corn flour and 30% corn starch (FF); GC: 20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch (GF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF); HC: 20% lupine flour, 30% rice flour and 30% corn starch (FF);

cookies. Blend AC contained the highest amount of protein (18.9%), ash (3.01%) and lipid (23.65%) since it was composed of 100% lupine flour. This result was expected since the proximate composition of lupine flour was the highest in protein, ash, fiber and lipid, as presented in Table 1. These results confirm the findings obtained by Zielinska *et al.* (2008) that lupine is a good source of nutrients, not only proteins but also lipids, dietary fiber, minerals and vitamins.

As for the carbohydrate content, this was highest in wheat flour cookies serving as a Control (WC) at 70.71%, but was also high in the blend DC compared to all blended flour cookies since it is composed of a large amount of corn starch (70%) and a low amount of lupine (30%). As for fiber, blend AC contained the highest amount of fiber, whereas blend FC contained the lowest amount. This substantial significant difference between blend AC and the others, as shown in Table 2, is due to the level of lupine flour presented in each blend. While blend AC was composed of 100% lupine flour, blend FC contained only 30% lupine. The protein content of the cookies was the highest in AC cookies (18.9%) compared with all blended flour cookies due to using 100% lupine flour. This was reduced in the other types of blended flour cookies and depended on the percentage of lupine flour in the formula.

Physical characteristics: The physical characteristics of the control and gluten-free cookies are shown in Table 3. The diameter of the blended flour cookies ranged 34.3 to 39.6 mm, whereas the highest diameter was for the Control (WC) at 40.8 mm. The thickness of the blended flour cookies ranged from 4.6 to 4.8 mm, whereas the smallest thickness observed was for the Control (WC) at 4.5 mm; Table 3. The spread factor values of the gluten-free cookies and control cookies ranged from 7.15-9.2. The spread value of all cookies type from different flours was affected significantly.

In general, all gluten-free cookies showed increased weight and thickness with a reduction in diameter and spreading.

These effects increased with an increasing level of wheat flour substituted with lupine flour which could be attributed to the water insoluble pentosans found in lupine; these may have been responsible for the reduction in spread factor (Pomeranz and Shellenberger, 1971).

Spread factor values showed that there was a negative correlation between the lupine flour level and the spread factor value. As demonstrated in blends AC-DC, as the lupine flour level decreased, so did the spread factor ratio. Several authors have found a negative correlation between cookie diameter and the protein content of the flour (Abboud *et al.*, 1985; Gaines, 1991; Kaldy *et al.*, 1993; Leon *et al.*, 1996).

Table 3: Physical characteristics of different cookie typ	bes
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Cookies type	Diameter	Thickness	Spread factor
WC	40.8 ± 0.42^{a}	$4.50\pm0.07^{\circ}$	$9.20{\pm}0.07^{a}$
AC	36.9 ± 0.40^{d}	4.70 ± 0.00^{ab}	7.87±0.24°
BC	37.6±0.14 ^{cd}	$4.80{\pm}0.00^{a}$	$7.85\pm0.07^{\circ}$
CC	34.3±0.07 ^e	$4.80{\pm}0.00^{a}$	7.15 ± 0.20^{d}
DC	37.8±0.35°	$4.80{\pm}0.00^{a}$	7.85±0.07 ^c
EC	39.9±0.21 ^b	4.65 ± 0.07^{b}	8.55 ± 0.07^{b}
FC	37.8±0.21°	4.70 ± 0.00^{ab}	$8.04{\pm}0.06^{\circ}$
GC	39.6±0.64 ^b	4.60 ± 0.00^{b}	8.65 ± 0.07^{b}
HC	37.9±0.21°	4.65±0.07 ^b	8.14±0.08 ^c

*: Values are the average of two replicates±the standard deviation; Different superscript letters in the same column indicate a significant ($p\leq0.05$) difference according to Duncan's test; WC: 100% wheat flour (control); AC: 100% Lupine Flour (LF); BC: 50% lupine flour and 50% corn starch (BF); CC: 40% lupine flour and 60% Corn starch (CF); DC: 30% lupine flour and 70% corn starch (DF); EC: 30% lupine flour, 40% rice flour and 30% corn starch (EF); FC: 30% lupine flour, 40% corn flour and 30% corn starch (FF); GC: 20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch (GF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (HF)

Table 4: Color characteristics of different cookie formulations

Cookie blends	L*	a*	b*
WC	71.07 ^b	-1.99 ^{bc}	35.49 ^d
AC	65.03 ^e	-1.28 ^{cd}	38.49 ^{cd}
BC	72.91 ^{ab}	-2.65 ^b	43.35 ^{ab}
CC	72.27 ^{ab}	-1.58°	41.41 ^{abc}
DC	71.67 ^{ab}	-2.67 ^b	36.64 ^{cd}
EC	73.71 ^a	-4.28 ^a	45.45 ^a
FC	68.33 ^{cd}	-1.68°	39.44 ^{cd}
GC	66.93 ^{de}	-4.04 ^a	36.86 ^{cd}
HC	70.10 ^c	-2.87 ^b	35.90 ^d

*: Values are the average of two replicates±the standard deviation; Different superscript letters in the same column indicate a significant ($p\leq0.05$) difference according to Duncan's test; WC: 100% wheat flour (control); AC: 100% Lupine Flour (LF); BC: 50% lupine flour and 50% corn starch (BF); CC: 40% lupine flour and 60% corn starch (CF); DC: 30% lupine flour and 70% corn starch (DF); EC: 30% lupine flour, 40% rice flour and 30% corn starch (EF); FC: 30% lupine flour, 40% corn flour and 30% corn starch (FF); GC: 20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch (GF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (HF)

Moreover, the composite flour apparently formed aggregates due to an increased number of hydrophilic sites found within the oligosaccharides, polysaccharides, water-soluble proteins and polar amino acids in lupine, corn and rice flour, which increased competition for the limited free water in the cookie dough (Hove, 1974; Cerning-Beroad and Filiatre, 1976).

Color measurements: The effects of the gluten-free cookie formulation on color measurements were assessed and are presented in Table 4. Namely, the top and bottom surface color of a baked cookie is a very important parameter in the initial acceptability of a tested product by consumers (Zucco *et al.*, 2011). According to Chevallier *et al.* (2000), the color of the cookie surfaces originates from non-enzymatic browning (Maillard reactions) between reducing sugars and amino acids as well as from starch dextrinization and sugar caramelization. The lightness (L*), redness

Cookie	Overall		Overall					
blends	impression	Overall flavor	texture	Overall color	Crust color	Crumb color	Hardness	Aftertaste
WC	7.80±1.13 ^a	7.70±1.34 ^a	7.20±1.75 ^a	$7.90{\pm}0.88^{a}$	2.70 ± 1.06^{b}	$2.40\pm0.52^{\circ}$	$2.40\pm0.84^{\circ}$	$1.00{\pm}0.00^{d}$
AC	1.40±0.52 ^e	1.40 ± 0.52^{e}	1.60±0.52 ^e	1.40±0.52 ^e	4.50±0.53 ^a	4.40 ± 0.52^{a}	4.50±0.53 ^a	5.00 ± 0.00^{a}
BC	8.30 ± 0.68^{a}	7.20±1.14 ^a	7.30±1.33 ^a	7.20±1.31 ^{ab}	2.70 ± 0.67^{b}	2.20±0.79°	3.50±0.53 ^b	2.50±1.08°
CC	5.60±1.07 ^b	5.90 ± 0.88^{b}	4.90±1.66 ^b	5.30±1.16°	2.50±0.53 ^{bc}	2.40±0.69°	3.60 ± 0.52^{b}	2.70±1.63°
DC	3.80±0.97°	4.50±1.35°	2.50±0.70 ^{de}	5.00±0.94°	1.70 ± 0.48^{d}	2.00±0.67°	3.50 ± 0.53^{b}	3.60±0.69 ^b
EC	3.10±0.88 ^{cd}	3.00 ± 0.82^{d}	3.90 ± 0.88^{bc}	6.50±1.43 ^b	1.90 ± 0.47^{cd}	2.30±0.67°	$3.90{\pm}0.88^{a}$	4.40 ± 0.52^{ab}
FC	2.30±1.25 ^d	2.30±1.16 ^{de}	2.40±1.07 ^{de}	2.30±1.16 ^e	4.30±0.67 ^a	$4.10{\pm}0.87^{a}$	$3.80{\pm}0.63^{b}$	4.00 ± 0.67^{b}
GC	$8.00{\pm}0.67^{a}$	$7.00{\pm}0.82^{a}$	7.70±0.95 ^a	7.70±0.82 ^a	2.40±0.52 ^{bc}	2.50±0.52 ^b	3.30±0.43 ^b	1.70 ± 0.67^{d}
HC	$2.80{\pm}0.79^{cd}$	3.10±0.99 ^d	3.20±0.03 ^{dc}	4.70±0.82°	$3.40{\pm}0.69^{b}$	3.10±0.74 ^b	$3.90{\pm}0.88^{ab}$	$3.90{\pm}0.88^{b}$

*: Values are the average of two replicates±the standard deviation; Different superscript letters in the same column indicate a significant ($p \le 0.05$) difference according to Duncan's test; WC: 100% wheat flour (control); AC: 100% Lupine Flour (LF); BC: 50% lupine flour and 50% corn starch (BF); CC: 40% lupine flour and 60% Corn starch (CF); DC: 30% lupine flour and 70% corn starch (DF); EC: 30% lupine flour, 40% rice flour, and 30% corn starch (EF); FC: 30% lupine flour, 40% rice flour, and 30% corn starch (GF); HC: 20% lupine flour, 30% rice flour, 20% corn flour and 30% corn starch (HF)

(a*) and yellowness (b*) values of gluten-free and control cookies showed that the ranges of cookie color for L*, a* and b* were 72.91 to 65.03-4.28 to -1.28 and 45.45 to 35.49, respectively. Blend EC (30% lupine flour, 40% rice flour and 30% corn starch) significantly (p \leq 0.05) received the highest L* value, while blend AC (100% lupine flour) received the lowest value, as it was darker. This indicated that the higher amount of lupine flour leads to a darker color.

Blend EC significantly ($p \le 0.05$) received the highest negative a* value, while blend AC received the lowest value. Blend EC (30% lupine flour, 40% rice flour and 30% corn starch) significantly received the highest b* value, while blend WC control received the lowest value. The difference in color characteristics may be attributed to the differences in colored pigment in the flours, which in turn depends on the biological origin of the plant (Singh *et al.*, 2003).

In addition, an increase in redness could be due to a high amount of proteins leading to increased interactions between reducing sugars and amino acids (Claughton and Pearce, 1989).

Organoleptic evaluation: The results of the sensory evaluation judged by panelists indicated that there were significant differences among blends for all attributes, as shown in Table 5. The quality score for the overall impression of cookies ranged from 1.4 for AC (100% lupine flour) to 8.3 for BC (50% lupine flour and 50% corn starch). WC (control), BC and GC significantly ($p\leq0.05$) received the highest overall impression scores compared to other types of cookies.

For other sensory attributes, i.e., overall flavor, overall texture and overall color, the BC and GC blends had the highest scores compared with all other blended flour cookies, while the other sensory attributes were significantly ($p \le 0.05$) different within all types of cookies. From the results of the sensory analysis (Table 5), blends BC (50% lupine flour and 60% corn starch) and GC (20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch) had the best sensory quality of liking attributes of all blends.

CONCLUSION

A basic formula and procedure for preparing gluten-free cookies was established. The principle materials were gluten-free flour blends (mainly lupine, corn and rice flour) and the function of gluten was performed by adding gums as gel-forming substances (xanthan and carrageenan). In this study, eight glutenfree flour blends were formulated with different flour compositions (lupine, rice, corn flour and corn starch). Under the test conditions studied, the cookies with the best overall quality were prepared with 50% lupine flour and 50% corn starch (blend BF) and blend GF that contained 20% lupine flour, 30% corn flour, 20% rice flour and 30% corn starch. The xanthan and carrageenan ratio (1:1) and the addition level of 1.5% (3.75 g) improved the rheological properties of the gluten-free dough (elasticity and cohesiveness). Lecithin at an addition level of 3% brought about extraordinary changes to the gluten-free dough (tenderness and softness). Therefore, this product could not only be used by celiac patients but also by patients with diabetes, cardiovascular disease and hypertension. This product could help reduce cholesterol and constipation as it is rich in fiber. Moreover, this product is a good source of prebiotics and thus helps increase the population of good micro-flora in the intestine. Finally, this product could be a major part of a weight reduction regimen since it contains high amounts of fiber and provides fewer calories.

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