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BIOLOGICAL, NUTRITIONAL AND SENSORY EVALUATION OF WHOLEGRAIN WHEAT BREADS SUPPLEMENTED WITH PURSLANE (PORTULACA OLERACEA) LEAVES

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ABSTRACT

Wholegrain wheat breads were prepared with the addition of high Purslane (*Portulaca oleracea*) Leaves at various levels (5%, 10% Flour basis). The breads made with the addition of *Portulaca oleracea* Leaves were sensorially acceptable, containing significantly more antioxidant, calcium and crude fibers. It was estimated that wholegrain supplemented breads would contribute reduction of :- cholesterol by 21.4%, 33.3% and 50.0%, triglycerides by 14.9%, 21.1% and 60.1% and LDL cholesterol by 39%, 69.8%, and 84.9% for rats fed on hyperlipidemic diet contain wholegrain only and wholegrain supplemented with 5%, 10% of *Portulaca oleracea* leaves respectively. While the addition of *Portulaca oleracea* leaves led to increase the content of calcium femur of rats fed on hyperlipidemic diet by 7.55% and 8.15% when the wholegrain supplemented with 5% and 10% of *Portulaca oleracea* leaves respectively.

Key Words: Wholegrain Bread – *Portulaca oleracea* – Sensory Evaluation – Rats

INTRODUCTION

Bread and bakery products have an important role in human nutrition. Generally, wheat bread is considered to be a good source of energy and irreplaceable nutrients for the human body. This is especially true for the products made from wholegrain or high-yield flour types. Bread prepared from refined flour is nutritionally much poorer and does not adequately meet the requirements for many macro- or micro-nutrients. It has been reported that bread made from

refined flour has low micronutrient content (Isserliyska et al., 2001). Also, wheat protein lacks the balance of essential amino acids- lysine, threonine and valine. Therefore, there have been many on-going investigations on enhancing the nutritive value of bread to fulfill the expanding demands of modern dietary habits, considering the products' protein, mineral, vitamin and/or fibre contents.

Phytic acid and phytase are the two major factors associated with the bioavailability of iron and calcium in wheat-based foods (Lott et al., 2000). Phytic acid is a strong chelator of mineral nutrient elements. The complex of phytic acid and mineral elements, in the form of phytate, leads to a marked reduction in bioavailability of these nutrient elements, and thus a consequent public health problem of iron and calcium deficiency for the populations whose diets are mainly cereals and legumes (Raboy, 2001).

Bakery products, supplemented with various nutritious, protective and ballast substances, have been gaining popularity worldwide. Mixed grain, wholegrain breads and related products are even considered as functional foods because they are convenient vehicles for important nutrients and phytochemicals (Biljana et al., 2008).

Portulaca oleracea L. (Portulacaceae) is a widespread weed. It is an edible plant and is used as a folk medicine in many countries, acting as a diuretic, febrifuge, antiseptic, antispasmodic or vermifuge as stated by Mohanapriya et al., (2006). It also has antibacterial, analgesic, anti-inflammatory, skeletal muscle relaxant, and wound-healing properties as mentioned by Lim and Quah, (2007). In addition to flavonoids, coumarins and monoterpene glycosides, alkaloids such as dopamine, dopa, noradrenaline, and betalains have been reported to be important chemical constituents of this plant. Betalains have attracted the interest of researchers in various fields owing to their antioxidant and radical-scavenging properties as reported by Chen et al., (2003). Recent research has shown that the *P. oleracea* is a rich source of calcium, iron, potassium, and omega-3 fatty acids, which is important in preventing heart attacks and strengthening the immune system (Simopoulos, 2004).

Therefore, the present investigation was undertaken to study whether supplementing wholegrain wheat breads with *Portulaca oleracea* Leaves at two levels of supplementation (5%, and 10%, Flour basis) would contribute to meeting the dietary requirements for

calcium and thus benefit the general population. Besides its effect on some physiological changes in rats such as body weight, weight of organs and food intake, biochemical changes including, total cholesterol, triglycerides, HDL, and LDL cholesterol, and the effect of adding the *Portulaca oleracea* on finished product sensory quality was also estimated.

MATERIALS AND METHODS

Purslane (*Portulaca oleracea*) Leaves:

They were obtained from the local markets, Egypt. They were cleaned and washed, then were dried at room temperature. After drying, *Portulaca oleracea* Leaves were homogenized to a fine powder and stored at room temperature. Powder was mixed with the flour at level of 5 & 10 %.

Table (1): Chemical composition of *Portulaca oleracea* leaves powder.

Composition	Content %
Total Carbohydrates	50
Fibers	11
Protein	26
Fat	4
Ash	9
Calcium	1.500
Iron	0.029
Potassium	1.800

Duke and Ayensu. (1985)

Wheat flour 72% extraction (14.6% moisture, 11.3% protein, and 0.61% ash) and wholemeal wheat flour (12.7% moisture, 1.72% ash content, 11.8% protein content), was obtained from South Cairo Flour Mills Co., Giza, Egypt.

Preparation of breads

The bread dough formula was: flour (refined or wholegrain) (100%), compressed yeast (3% for breads made with refined flour, and 4% for breads made with wholegrain flour), salt (2%), gluten (2% for breads made with refined flour, and 4% for breads made with wholegrain flour), commercial improver (dosed as recommended by the manufacturer), water (optimum), and *Portulaca oleracea* Leaves

(tested at 5% and 10% levels). Percentages are based on flour weight. Bake trials were carried out under laboratory conditions. Dough mixing, processing and baking were performed on laboratory-scale equipment. A straight dough process was used. Doughs were mixed to optimum consistency in a high-speed mixer with low speed of 85 r/min for 1 min and high speed of 120 r/min for 7 min. Final dough temperature was 30°C. Doughs rested in bulk for a period of 45 min, and then they were hand-kneaded and left to rest for 15 min. Doughs were scaled into 340 g portions, manually rounded, rolled, put into tin pans (24.5 X 9 X 6.5 cm) to final fermentation for 75 min (for bread made with refined flour), i.e. 40 min (for bread made with wholegrain flour) at 30°C and 80% relative humidity. Baking was done at 230°C in a deck-type oven until the initial dough mass was reduced by 8%. (Biljana et al., 2008)

Bread evaluation

Bread quality parameters included weight, volume (determined by seed displacement in a loaf volume meter), specific volume, acceptance and texture of crumb. Overall acceptability was carried out as follow: one slice of bread, identified by code numbers, was served to each panelist (10 panelists) under normal (daylight) illumination. They evaluated each product for quality attributes: Color, grain, crumb smoothness, aroma, flavor and overall acceptability. Acceptability of each quality attribute was rated with score 1 (lowest) to 10 (highest). Products were considered acceptable if their mean scores of overall acceptability were 5 (neither like nor dislike) (Wang et al., 2002).

Experimental animals:

Twenty five male of white albino rats (Sprague dawleey strain) weighting (100-115 gm) were obtained from National Research center. Dokki, Egypt. They were housed in individual cages at room temperature. The animals were left to acclimatize for ten days before the start of experiment. They were fed with standard laboratory diet according to AOAC, (1995).

Experimental design:

The biological experiment was designed to study the effect of wholegrain bread with or without *Portulaca oleracea* Leaves on biochemical effects on serum cholesterol, triglyceride, HDL, LDL,

body and organs weight, and calcium femur content for hyperglycemic rats.

The rats were divided into 5 groups each of 5 rats as follows:

Group 1: Negative Control group fed on standard diet

Group 2: Positive Control group fed on hyperlipidemic diet

Group 3: fed on hyperlipidemic diet with 15% wholegrain bread

Group 4: fed on hyperlipidemic diet with 15% wholegrain bread supplemented with 5% PO Leaves

Group 5: fed on hyperlipidemic diet with 15% wholegrain bread supplemented with 10% PO Leaves

Experimental period for all groups was 40 days. Animals were weighted weekly and sacrificed at the end of the experiment. Blood samples were collected from hepatic portal vein; serum was separated by centrifugation at 3000 rpm, for 15 minutes and kept at -5 °C till analysis. Liver, kidney, heart and spleen were removed and kept in formalin at room temperature.

Table (2): The composition of group diets used in this investigation.

Ingredients	G1	G 2	G3	G4	G5
Corn Starch	67	67	52	52	52
Casien	13	13	13	13	13
Corn oil	10	-	-	-	-
Vit Mix	1	1	1	1	1
Salt Mix	3.5	3.5	3.5	3.5	3.5
Cellulose	5	5	5	5	5
Choline Chloride	0.2	0.2	0.2	0.2	0.2
Animal fat	-	10	10	10	10
Cholesterol	-	1	1	1	1
wholegrain Bread	-	-	15	14.25	13.5
Portulaca Oleracea	-	-	-	0.75	1.5

Biochemical analysis:

Samples of blood was taken and centrifuged (3000 rpm/10 min.) to obtain serum. Serum was used to determine total cholesterol

(Roeschlau et al., 1974), triglycerides (Lowell and Randal, 1973), high density lipoprotein, low density lipoprotein (Allian et al., 1974). Calcium femur content was assayed according to the methods of Platt et al. (1987) and Platt and Clydesdale (1986).

Statistical analysis

The statistical analyses were conducted using one-way ANOVA procedures. Differences in samples due to the addition of *Portulaca oleracea* were tested for statistical significance at the $p = 0.05$ level. Tukey's Honestly Significant Difference was used to differentiate between the mean values. Calculations were performed using SPSS version 16.0 (SPSS, Chicago, IL, USA).

RESULTS AND DISCUSSION

Sensory Evaluation

Sensory evaluation of breads (Table 3) implied that, the control get the highest score in all parameters than wholegrain bread and wholegrain supplemented with *Portulaca oleracea* leaves, as the level of *Portulaca oleracea* leaves increased in wholegrain bread, color, grain, and crumb smoothness decreased, while the Aroma increased. The Flavor was not significantly affected by the addition of *Portulaca oleracea* as compared to the corresponding control sample. The addition of *Portulaca oleracea* improved the aroma of bread samples. The specific volume of all wholegrain breads decreased, but not significantly, with the addition of *Portulaca oleracea*. The specific volume of wholegrain breads was significantly lower than that of white breads. The addition of *Portulaca oleracea* generally affected the shapes of wholegrain breads giving flatter products. These changes were less pronounced for wholegrain bread types where only the sample with the highest supplementation level differed significantly from the corresponding control.

Table (3): Sensory and Physical characteristics of Bread from refined wheat flour and it supplemented with various levels of *Portulaca oleracea* (5 & 10%).

Parameters	White Bread (Control)	Wholegrain Bread	P O leaves %	
			5.0	10.0
Loaf volume (ml)	175	112	106	89
Loaf weight (g)	31	31	31	31
Specific volume (ml/g)	5.64	3.62	3.43	2.88
Sensory analysis				
Color	9.5 ^a	7.8 ^b	7.4 ^{bc}	6.3 ^c
Grain	9.2 ^a	8.6 ^b	7.5 ^b	6.4 ^c
Crumb smoothness	9.5 ^a	7.6 ^b	6.8 ^c	5.8 ^d
Aroma	9.0 ^a	8.5 ^b	8.7 ^a	8.9 ^a
Flavour	9.6 ^a	9.0 ^b	8.7 ^{ab}	8.8 ^{ab}
Overall acceptability	9.4 ^a	8.5 ^b	7.2 ^{bc}	6.1 ^c

Means with the same letter are insignificantly different.

* Control is referred to Bread without added leaves.

** Nine point hedonic scale ratings: 9= like extremely and 1=dislike extremely

Feed intake and body weight gain

Figure (1) shows mean values of daily feed intake, body weight gain, daily body weight gain and feed conversion ratio of normal and hyperlipidemic rats fed wholegrain bread supplemented with different levels (5 & 10%) of *Portulaca oleracea* leaves. There is no significant difference between the daily feed intake of normal (11.91 ± 0.28 g/d/rat) and all groups except group (3) which fed on wholegrain bread only (11.27 ± 0.19 g/d/rat), but there are significant differences ($P < 0.05$) between those fed wholegrain with 5% *Portulaca oleracea* leaves (12.18 ± 0.40 g/d/rat) and those fed on wholegrain only and untreated group. While there is no significant difference between rats fed on wholegrain with 10% *Portulaca oleracea* leaves (11.77 ± 0.61 g/d/rat) and those fed on wholegrain only, wholegrain with 5% *Portulaca oleracea* leaves and untreated group.

The higher value in body weight gain per day of untreated group (3.45 ± 0.27 g/d/rat) than other groups as show in Figure (1), due to the

interrelation between dietary fat specially saturated fatty acids and obesity Huang et al., (2004).

There is no significant difference between untreated group and normal group for body weight gain per day or feed conversion, but the untreated group increased significantly than other groups for body weight gain per day and decreased significantly than other groups for feed conversion.

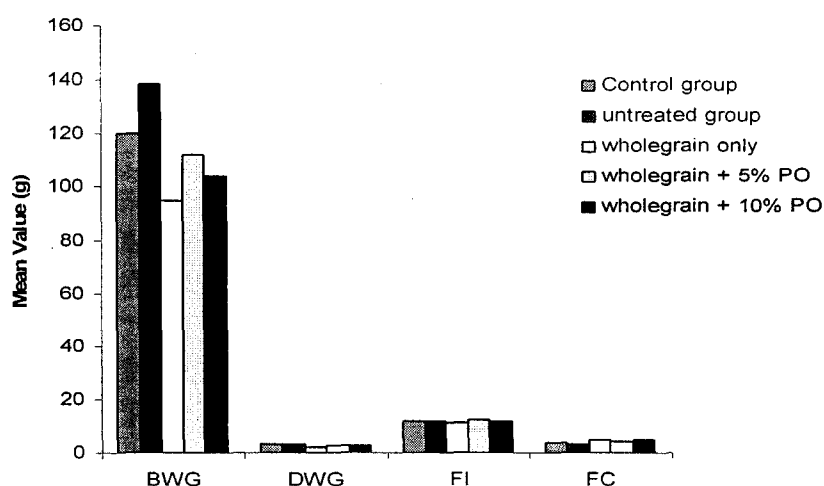


Figure (1): Mean value of feed intake (FI), body weight gain (BWG), daily body weight gain (DWG) and feed conversion (FC) ratio of hyperlipidemic rats fed wholegrain bread supplemented with different levels (5,10%) of *Portulaca oleracea* leaves

Relative weight of Organs

Figure (2) demonstrates the relative weight of some organs of normal and hyperlipidemic rats fed wholegrain bread supplemented with different levels (5 & 10%) of *Portulaca oleracea* leaves.

There is highly significant differences between control groups and untreated rats, but less significant with those fed on wholegrain only. While no significant differences between the control and other groups. The addition of *Portulaca oleracea* leaves caused decrease in liver weight may be due to the effect of some natural antioxidants in *Portulaca oleracea* leaves which made reduction in fat content in the liver (Lim and Quah, 2007)

About the relative weight of kidney, the highest value was (0.49 ± 0.054) for untreated group, it decreased significantly to (0.46 ± 0.034) , (0.44 ± 0.079) , and (0.36 ± 0.026) for those fed on wholegrain with 10%, 5% and wholegrain only respectively. No significant between untreated group and other group except group (3) which fed on wholegrain only.

There is significant difference between the relative weight of spleen for control group and untreated group, but no significant with other groups. About the relative weight of heart there is no significant difference between groups.

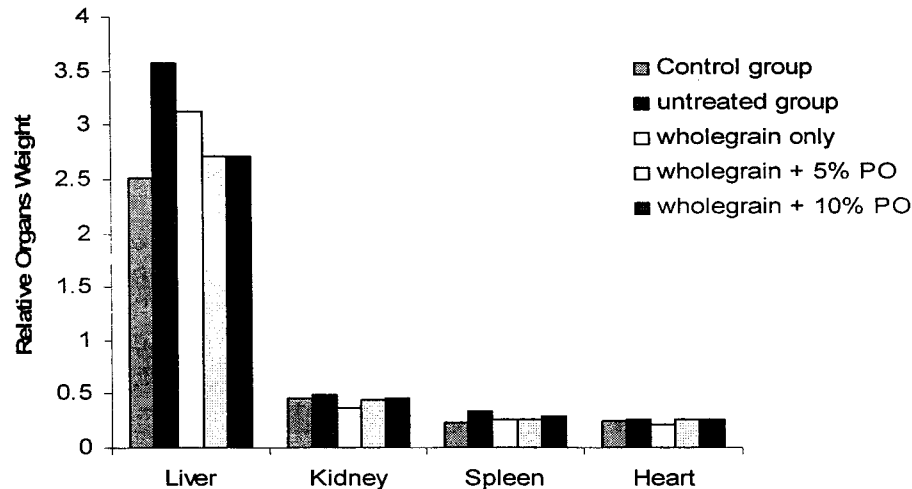


Figure (2): Mean value of relative organs weight of hyperlipidemic rats fed wholegrain bread supplemented with different levels (5 & 10%) of *Portulaca oleracea* leaves

Biological parameters

The serum of total cholesterol, triglyceride, HDL, LDL, and Calcium femur content of normal and Hyperlipidemic rats fed on wholegrain bread only or supplemented with different levels (510%) of *Portulaca oleracea* leaves were recorded in table (4).

The data in table 4 showed that, there is highly significant difference for reduction in serum total cholesterol between untreated group (140.76 ± 47.66 mg/dl) and all groups of animals except those fed wholegrain only (110.63 ± 23.78 mg/dl). While there is no significant difference between control group and each of those fed wholegrain supplemented with 5% and 10% of *Portulaca oleracea* leaves. This reduction of cholesterol equal 21.4%, 33.3% and 50.0% for those fed wholegrain only and wholegrain supplemented with 5% & 10% of *Portulaca oleracea* leaves respectively. This result agreed with Simopoulos (1995) who reported that, *Portulaca oleracea*, contain protective factors against chronic disease and their influence on cholesterol metabolism as lowering effect. Dried *Portulaca oleracea*, contains large quantities of $\omega 3$ polyunsaturated fatty acids, that recommended as cholesterol lowering agents (Koch, 1988).

The data in table (4) showed that serum triglycerides (176.54 ± 47.79 mg/dl) for untreated group decreased significantly with adding 10% of *Portulaca oleracea* and reached to 70.38 ± 10.88 mg/dl (decreasing ratio 60.1%), but non significant with adding 5% *Portulaca oleracea* leaves 139.28 ± 20.21 mg/dl (decreasing ratio 21.1%) and wholegrain only 150.27 ± 70.51 mg/dl (decreasing ratio 14.9%).

The reduction of triglycerides level may be due to the inhibitory effect of wheat and *Portulaca oleracea* fibers as suggested by Simopoulos, (1995) who found that, *Portulaca oleracea* contain protective factors that influences as lowering effect on blood lipid levels in human and rats. Also Aline Adam et al., (2003) found that the apparent cholesterol absorption related to dietary cholesterol intake was significantly reduced in Whole Wheat Bread-fed rats compared to the control.

There is no significant difference between untreated group and all groups for HDL cholesterol. While there is highly significantly difference between untreated group and all groups except those fed on wholegrain only for LDL cholesterol. The highest value of LDL cholesterol was 72.26 ± 41.74 mg/dl for untreated group then, it

decreased significantly by treatment to 44.03 ± 23.54 mg/dl (39%), 21.81 ± 18.03 mg/dl (69.8%), and 10.92 ± 2.53 mg/dl (84.9%) for rats fed hyperlipidemic diet contain, wholegrain only, wholegrain supplemented with 5% and 10% *Portulaca oleracea* leaves, respectively. The hypocholesterolemic effect of plant occur by increasing of HDL cholesterol level which to increase in the follow of cholesterol to liver converted to other compounds (Ibrahim, 1997).

Regarding to calcium femur content, there is significant difference between group (3) (283.25 ± 23.39 mg/dl) which fed on wholegrain only and each of those fed on wholegrain supplemented with 5% (304.64 ± 10.82 mg/dl) with increasing ratio 7.55%; and 10% *Portulaca oleracea* leaves (306.34 ± 9.74 mg/dl) with increasing ratio 8.15%. But there are no significant differences between other groups. The addition of *Portulaca oleracea* leaves led to increase of calcium content and lowered the reduction of calcium which resulted from the effect of phytic acid in wholegrain bread, and it may be due to its rich in calcium (Simopoulos, 2004).

Table (4): Serum total cholesterol, triglyceride, HDL, LDL, HDL/LDL ratio and Calcium femur content of hyperlipidemic rats fed high fiber bread supplemented with different levels (5,10%) of P.O. leaves.

Groups	Total Cholesterol Mg/dl	Total Triglyceride Mg/dl	HDL Mg/dl	LDL Mg/dl	HDL/LDL	Calcium Content Mg/dl
1	69.18 ± 9.10	69.02 ± 24.33	46.75 ± 8.91	8.63 ± 4.33	7.57 ± 5.71	297.55 ± 7.52
2	140.76 ± 47.66	176.54 ± 47.79	36.93 ± 15.91	72.26 ± 41.74	0.71 ± 0.50	290.88 ± 13.24
3	110.63 ± 23.78	150.27 ± 70.51	38.44 ± 4.18	44.03 ± 23.54	1.02 ± 0.40	283.25 ± 23.39
4	93.87 ± 23.02	139.28 ± 20.21	38.69 ± 5.97	21.81 ± 18.03	2.38 ± 1.05	304.64 ± 10.82
5	70.38 ± 10.88	67.87 ± 32.25	43.51 ± 7.29	10.92 ± 2.53	4.28 ± 1.72	306.34 ± 9.74

Conclusion

From the present paper it may be inferred that *Portulaca oleracea* leaves could be added to wholegrain bread up to levels of 5% (Flour basis) without significant adverse effects regarding the crust color, Aroma, Flavor and Overall acceptability. Adding *Portulaca oleracea*

leaves significantly decreases crumb smoothness compared to the control samples but not to the level that would disqualify the product. The breads containing *Portulaca oleracea* leaves at all levels were scored higher for Aroma in comparison to the wholegrain breads. Besides sensory properties, *Portulaca oleracea* leaves supplemented breads were more acceptable in many nutritional aspects as they contained significantly more antioxidants, calcium and crude fibre. Such breads may contribute to enhancement of calcium status of certain population groups since dietary surveys suggest that many individuals (particularly aged people or those taking medications such as hormone replacements or diuretics) fail to achieve the dietary reference value for calcium. According to data from this investigation, the addition of *Portulaca oleracea* leaves caused decrease in liver weight due to reduction in fat content in the liver, reduction in total cholesterol, triglycerides and LDL cholesterol. Also the addition of *Portulaca oleracea* leaves increased calcium content. In any case, consumption of breads supplemented with *Portulaca oleracea* leaves could be beneficial for improving the nutritional and physiological status of the general population since Egypt is a poor developing country with high bread consumption.

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التقييم البيولوجي والتغذوي والحسي لخبز القمح الكامل المدعم بأوراق الرجلة

عبدالرحمن رجب عبدالرحمن

مدرس التغذية وعلوم الأغذية بقسم الاقتصاد المنزلي بكلية التربية النوعية جامعة عين شمس

تهدف الدراسة الي زيادة محتوى الكالسيوم في خبز القمح الكامل نظرا لما يفقد من هذا العنصر لوجود حمض النيك الموجود في الاغلفة الخارجية للقمح ونظرا لمحتوي أوراق الرجلة العالي من الكالسيوم فضلا عن تأثيرها الفسيولوجي في الجسم كما تبين ذلك من الدراسات السابقة فإنه تم تدعيم خبز القمح الكامل (المرتفع في نسبة الالياف) بإضافة مطحون أوراق الرجلة بمستويات مختلفة (5%، 10% علي أساس وزن الدقيق). وقد أوضحت الدراسة أن الخبز المدعم بأوراق الرجلة كان مقبول حسيًا، كما أنه يحتوي بكميات معنوية على الكالسيوم ومضادات الأكسدة، والألياف الغذائية. كما بينت الدراسة البيولوجية أن تغذية الفئران علي خبز قمح كامل مدعم بأوراق الرجلة قد أدت الي :- خفض نسبة الكوليسترول الكلي بنسبة 21.4%، 33.3% و 50.0%، الجليسيريدات الثلاثية بنسبة 14.9%، 21.1% و 60.1% و الكوليسترول الضار قليل الكثافة بنسبة 39% و 69.8% و 84.9% للفئران التي تغذت علي غذاء عالي في نسبة الدهون يحتوي علي، خبز قمح كامل فقط و خبز قمح كامل مدعم بنسبة 5%، 10% من أوراق الرجلة على التوالي. وقد لوحظ أيضا، أن إضافة أوراق الرجلة أدت الي زيادة محتوى الكالسيوم بنسبة 7.55% و 8.15% في عظمة فخذ الفئران المغذاة علي غذاء عالي في نسبة الدهون يحتوي علي خبز قمح كامل مدعم بنسبة 5%، 10% من أوراق الرجلة على التوالي. لذلك تنصح الدراسة بتدعيم خبز القمح الكامل بمطحون أوراق الرجلة بنسبة 5% لرفع نسبة الكالسيوم وتقليل نسبة الكوليسترول الضار في الجسم علما بأن هذه النسبة ذات تأثير غير معنوي علي الصفات الحسية للخبز الناتج.