The Potential Effects of Garden Cress Seeds *(Lepidium Sativum L.)* on the Bone of Female Rats Suffering from Osteoporosis

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Abstract

Osteoporosis is associated with a high rate of morbidity and mortality worldwide. This study was designed to investigate the probable effect of Garden Cress seeds (GCs) on the bone of osteoporotic female rats. Thirty healthy adult female albino rats (weigh, 200±10g) were used in this study. The rats were haphazardly divided into two main groups. The first main group (6 rats) was fed on a basal diet as a control negative group. The second main group (24 rats) fed on a basal diet containing 100mg prednisone acetate as a source of glucocorticoid/kg diet for two weeks to induce osteoporosis. One group was selected as a positive control group, three groups were fed on prednisone acetate diets containing GCs powder at the level of 2.5, 5, and 7.5%. The proximate chemical composition of GCs powder contained 2.75, 23.67, 22.81, 5.16, 8.42, and 37.19 g/100g for moisture, protein, fat, ash, crude fiber, and total carbohydrates respectively, while caloric value recorded 448.73 Kcal/100g. The minerals content were 309.21, 612.54, 341.10, and 5.74 mg/100g for calcium, phosphorus, magnesium, and zinc. The biological results showed a significant improvement ($P \leq 0.05$) in values of Ca and P in serum and femur bone of osteoporotic rats fed on a basal diet containing different ratios of GCs. Bone mineral content and bone mineral density also increased as compared to the control positive group. Kidney and liver functions significantly (P≤0.05) improved compared to the positive control group. The histological examination of bone confirmed a gradual improvement in all treated groups. Peanut sweets were produced with proportions 2.5, 5, and 7.5% of GCs, sensory evaluation indicated that all peanut sweets samples have an acceptance greater than 75%. In conclusion, the current study indicates that GCs have improved osteoporosis biomarkers in rats and can be used up to 7.5% as a supplement in the diet of osteoporosis patients.

Keywords: osteoporosis**,** peanut sweets, calcium, phosphorus, bone mineral density.

Introduction

Osteoporosis is a chronic metabolic bone disease characterized by low bone mineral density (BMD), degradation of the bone structural, and increased fracture risk **(Wang et al., 2017).** Women over 45 years of age are the most vulnerable to femoral fractures, which are rising and pose a health and economic burden on society **(Wu et al., 2018).** Recent treatments of osteoporosis concentrate on slowing bone resorption and increasing bone remodeling **(Wu et al., 2017).** Pharmacological treatments that including calcium and vitamin D supplementation have been associated with a variety of side effects, especially with long-term usage **(Hough et al., 2014).**

Natural plant products as medication substitutes remain an important element of traditional medical regimens in developing nations **(Veeresham, 2012).** Garden cress *(Lepidium sativum L.)* is a fast-growing annual herb from the family *Cruciferae*, native to Egypt and west of Asia and is presently cultivated all over the world **(Singh *et al.*, 2015)**. Garden cress seeds (GCs) are an edible plant known as “Hab Al-Rashad” in Egypt, it can be used to fortify a variety of beverages and foods due to its excellent nutritional and functional qualities **(Sakran *et al.*, 2014)**.

GCs include 27% protein, fat ranging from 14 to 26%, carbohydrates from 35 to 54%, and 8% crude fiber. GCs carbohydrates are composed of 10% starch and 90% non-starch polysaccharides, GCs contain 20–25% oil, with α-linolenic acid accounting for 32–35% of the totality, while the total essential amino acid content is 47.08% **(Chatoui *et al.*, 2020).** They also include natural antioxidants that aid in the protection from oxidation and rancidity in the oil **(Zia-Ul- Haq *et al.*, 2012).** GC seeds have a remarkable amount of iron, calcium, phosphorus, folic acid, vitamins C and A **(Lahiri and Rani, 2020)**. Furthermore, the phytochemicals ingredients in seeds contain alkaloids, saponins, and flavonoids **(Manohar *et al*, 2012).**

GCs are used in several medicinal applications as a tonic for the immune system, in treating anemia **(Doke and Guha, 2014),** antidiarrheal **(Manohar *et al.*, 2009),** antidiabetic **(Rahimi 2015),** antihypertensive, diuretic **(Maghrani *et al.*, 2005)**, antioxidant (**Yadav 2010),** anti-carcinogenic **(Al-Sheddi *et al.*, 2016),** anti-hypercholesterolemia **(Al Hamedan 2010),** antibacterial, antifungal, anti-inflammatory **(Alqahtani *et al.*, 2019),** anti-osteoporosis activity**,** normal contraction of the muscles for healthy of limbs and heart activity **(Jabeen *et al.*, 2017),** and accelerate fracture healing **(Abdallah *et al.*, 2020 and Shah *et al.*, 2021)**.

Moreover, GCs have a potential therapeutic effect against liver toxicity and hepato-carcinoma **(Ahmad *et al.*, 2020),** also used in lactating mother's diets to enhance milk secretion during lactation, as well as to help regulate menstruation and thyroid hormone concentration **(Obeid, 2020 and Sciarrillo *et al.*, 2018).** Additionally, **Mabrouk *et al.* (2020)** discovered that GCs oil improved the osteoporosis bone marker in both therapeutic and preventative methods. According to **Datta *et al.* (2011),** GCs did not cause toxic effects even at 10% dietary level, as shown by the absence of any negative effects on growth or analytical parameters which means GCs are safe to consume in nutritional compositions.

Many people are still ignorant of the benefit of consuming GCs in their diets. Therefore, the present study aims to investigate the potential effect of GCs on the bone of osteoporotic rats, in addition to evaluating the sensory characteristics of peanut sweets supplemented with Garden cress seeds.

Materials and Methods

Materials

- Garden cress seeds (GCs) were purchased from Haraz Company for Agricultural Seeds, Perfumery and Medicinal Plants, Egypt.
- Casein, vitamins, minerals, cellulose, and choline chloride were purchased from El-Gomhoriya Company for Trading Drugs, Chemicals and Medical instruments, Cairo, Egypt.
- Prednisone acetate was purchase from a pharmacy in Damietta Governorate, Egypt.
- All ingredients used in peanut sweets formulation (Peanut seeds, sugar, corn syrup, vanilla flavor, and lemon) were obtained from the local market from Damietta Governorate, Egypt.

Methods

Preparation of Garden cress seeds

GCs were cleaned manually to remove adhering dirt, dust, and foreign particles along with shrunken and broken seeds. The seeds were roasted in a preheated iron skillet at a temperature of 128°C for 3 minutes leading to loss of raw flavor and production of a prominent aroma **(Vaishnavi and Gupta, 2020).** Roasted seeds were taken out from the skillet and spread on a tray for rapid cooling. After cooling, GCS were ground in electric milling. Packaging of roasted seeds flour was in air-tight plastic bags and stored in an air-sealed container till use.

Determination of gross chemical composition

The proximate major chemical constituents; moisture, ash, total protein, crude fiber, and fat in GCs powder were determined according to **A.O.A.C. (2000),** while total carbohydrates were calculated by differences. Caloric value was calculated according the following equation: Caloric value = 4 (protein%+ carbohydrates%) + 9 (fat%).

Minerals content in GCs; calcium, phosphorus, magnesium, and zinc were measured using Perkin Elmer Atomic Absorption Spectrophotometer (Model 2380, Japan) according to the method of **A.O.A.C. (2010)**

Biological experiments design

The biological studies were carried out in accordance with the National Research Council's Institute of Laboratory Animal Resources, Commission on Life Sciences regulations **(NRC, 2011).** Thirty healthy adult female albino rats Sprague Dawley Strain weighing 200±10g per each were purchased from the Laboratory Animal Unit in the Nile Center of Experimental Research, Mansoura, Egypt. The rats were individually housed in wire cages under the normal laboratory condition and were fed on a basal diet for one week as an adaptation period. The basic diet was prepared according to the following formula as mentioned by **(AIN, 1993),** while the used vitamins and salt mixtures components were formulated according to **Campbell, (1963)** and **Hegsted *et al.* (1941)**, respectively. After that, rats were randomly divided into two main groups, the first main group (6 rats) was fed on a basal diet, as a control negative group (C-). The second main group (24 rats) fed on a basal diet containing 100mg (prednisone acetate) as a source of glucocorticoid / kg diet for two weeks to induce osteoporosis according to **Liao *et al.* (2003)** The second main group was divided into four subgroups of 5 rats each as follows; subgroup 1: positive control (C+) fed on a prednisone acetate diet, subgroup 2: fed on a prednisone acetate diet containing GCs 2.5%, subgroup 3: fed on a prednisone acetate diet containing GCs 5%, and subgroup 4: fed on a prednisone acetate diet containing GCs 7.5%. Rats were maintained under standard conditions (23±2°C temperature, 55±5% relative humidity, 12h light/12h dark cycle). The animals were fed diet and water ad-libitum for a period of 4 weeks. The diets consumed as well as body weights were recorded twice weekly.

At the end of the experiments, all rats were fasted up to 12 hours and then sacrificed. Blood samples were collected from the aorta. The blood samples were centrifuged, separated, and stored frozen at -20°C until further analysis.

Biochemical evaluation

Calcium and Phosphorus: Serum calcium and phosphorus samples were analyzed according to **Baginski (1973)** and **Yee (1968),** respectively. Calcium and phosphorus in the femur bone of the rats were determined according to the method described by **Muynck and Vanhaecke, (2009).**

Kidney functions: Serum levels of uric acid, urea nitrogen and creatinine were determined according to **Fossati *et al.* (1980), Patton and Crouch (1977),** and **Bohmer (1971),** respectively.

Liver functions: Serum levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities were measured according to **Tietz (1976),** while Alkaline phosphates (ALP) activity was determined according to **Vassault *et al.* (1999).**

Bone Mineral Density (BMD) and Bone Mineral Concentration (BMC) were both measured by Dual Energy X-ray Absorptiometry (DEXA) in the osteoporosis unit at the Egyptian Center for Medical and Scientific Research of Excellence.

Histological examination

Each rat's bones were removed, cleaned, weighed and specimens of right cortical bone were fixed with 10% neutral formalin solution till histopathologically examined according to **Bancroft and Cook, (1998).**

Preparation of Peanut Sweets

Preparation of standard formula from peanut sweets (control) was produced by following the traditional method; Peanuts (100g), sugar (50g), corn syrup (25g), water (20ml), and lemon juice (5ml). Sugar and water were heated and made into syrup, then lemon juice was added while stirring the mixture till the temperature reached (150°C). The mixture was left boiling till the syrup turned glossy and thickened and the hard crack stage was reached. The corn syrup was added and the mixture stirred until completely homogeneous, then flavored with vanilla and kept away from the flame. Roasted and crushed peanuts were added to this syrup and were mixed thoroughly to cover the peanuts properly. The mixture was then transferred on a pre-greased surface and spread uniformly with the help of a roller, cut into square pieces, and put away to cool at room temperature. The other experimental formulations were prepared by addition of powder from GC seeds with 2.5, 5, and 7.5%.

Sensory analysis

Peanut sweets samples were served in random order to ten panelists of staff members from Damietta University, Damietta, Egypt. Sensory attributes for color, taste, flavor, texture, appearance, and acceptance were evaluated using a 9- point hedonic scale (1= dislike extremely to 9 = like extremely) **(IFT, 1981).** Water was served for cleaning the mouth between samples.

Statistical Analysis:

Data obtained were statistically analyzed by SPSS computer software SPSS 2000. The results were expressed as mean ± standard deviation (SD) and tested for significance using one-way analysis of variance ANOVA test, according to **Armitage and Berry, (1987).**

Results and Discussion

Proximate chemical composition of Garden cress seeds

The proximate chemical composition of Garden cress seeds powder (GCs) was investigated on a dry weight basis and presented in table (1). It could be observed that GCs contained 2.75±0.09, 23.67±0.05, 22.81±0.06, 5.16±0.13, 8.42±0.07, and 37.19 g/100g for moisture, crude protein, fat, ash, crude fiber, and total carbohydrate, respectively. These findings revealed that seeds had a high amount of protein and low moisture content that is an indicator of stable quality and increased shelf life of seeds. Additionally, GCs had a high food energy content that reached 448.73 Kcal/100g for caloric value. On the other hand, GCs contained several minerals; 309.21±0.14, 612.54±0.12, 341.10±0.06, and 5.74±0.03 mg for calcium, phosphorus, magnesium and zinc, respectively. The results showed that GCs are a good source of minerals that can protect bones.

Table (1): Proximate chemical composition of GCs powder (%On dry weight basis)

Components	Ingredients
Moisture (g)	2.75 ±0.09
Protein (g)	23.67±0.05
Fat (g)	22.81±0.06
Ash (g)	5.16±0.13
Crude fiber (g)	8.42±0.07
Total Carbohydrate (g)	37.19
Caloric Value (Kcal)	448.73
Calcium (mg)	309.21±0.14
Phosphorus (mg)	612.54±0.12
Magnesium (mg)	341.10±0.06
Zinc (mg)	5.74±0.03

Each value represents the mean ± SD.

Feed intake, body weight gain% and femur bone weight relative to body weight %

Results of feed intake (FI), body weight gain% (BWG%), and femur bone weight/body weight% are summarized in table (2). It could be observed that FI (g/day/rat), BWG%, and femur bone weight/body weight% of the osteoporotic group (C+) decreased significantly ($p \leq 0.05$) when compared to the control negative group. That was due to the rats receiving a basal diet containing prednisone acetate, which reduced both appetite and body weight. Osteoporotic groups which fed on a prednisone acetate diet containing 2.5%, 5%, and 7.5% GCs showed a high gradual increase in FI, BWG%, and femur bone weight/body weight% as compared to the control (+) group. Statistical analysis showed significant positive correlations between treatments for FI, BWG%, and femur bone weight/body weight% by GCs. These results are in line with those reported by **Mali *et al.* (2007)** who found that *Lepidium Sativum* contains growth promoter factors as triterpenes, alkaloid, tannin, and coumarins. **Al Hamedan (2010)** indicated that rat groups with oral administration of garden cress seed extract showed a significantly lower value of weight gain.

Table (2): Effect of GCs on feed intake, Body weight gain% and femur bone weight/body weight% of female osteoporotic rats

Parameters / Groups	Feed Intake g/day/rat	BWG%	Femur Weight/ body weight%
Control -	17.34[a] ±1.56	20.70[a] ±1.78	1.63[a] ±0.07
Control +	11.21[c] ±0.85	7.53[d] ±1.22	0.84[d] ±0.09
2.5% GCs	13.54[b] ±1.15	9.14[c] ±1.83	1.10[c] ±0.06
5% GCs	14.32[b] ±0.53	12.31[b] ±1.69	1.28[b] ±0.05
7.5% GCs	14.71[b] ±1.21	13.83[b] ±1.70	1.32[b] ±0.06

GCs: Garden cress seeds
Means in the same column with different superscript letters are significantly different at $p \leq 0.05$.

Serum calcium and phosphorus, Femur bone calcium and phosphorus, bone mineral density, and bone mineral concentration

Calcium is the major component of the bone skeleton and possesses a significant role in osteoporosis prevention because its level is directly related to bone mineral density (BMD) and bone health **(Hejaz et al., 2020).** The calcium-to-phosphorus ratio, rather than the calcium or phosphorus level alone, is a deciding factor in bone validity and an osteoporosis prediction **(Shakoor et al., 2014).**

Results of feeding osteoporotic female rats on diets containing different levels of GC seeds on serum calcium and phosphorus, femur bone calcium and phosphorus, bone mineral density (BMD), and bone mineral concentration (BMC) were illustrated in the table (3). It could be noticed that untreated osteoporotic group (C+) recorded significant decreases in serum calcium and phosphorus (mmol/l) as compared them with the healthy control group (C-) (2.19±0.21 and 1.42±0.15 vs. 5.04±0.31 and 3.12±0.37 mmol/l, respectively), Treating osteoporotic groups with a prednisone acetate diet supplemented with 2.5, 5, and 7.5% GCs improved the levels of serum calcium and phosphorus in rats suffering from osteoporosis. The highest increase in serum calcium and phosphorus levels appeared in the osteoporotic group fed on a prednisone acetate diet supplemented with 7.5% of GCs which recorded non-significant changes as compared to the control negative group.

Femur bone calcium and phosphorus of healthy rats (C-) fed on a basal diet increased than that of osteoporotic rats (C+) (50.2±2.92 and 94.64±2.50 vs. 30.62±3.03 and 61.36±4.66 mg/kg, respectively). Treating osteoporotic groups with a prednisone acetate diet supplemented with 5% and 7.5% GCs elevated the levels of calcium and phosphorus in femur bone of rats suffering from osteoporosis. The best result was recorded for the group of female rats which received a prednisone acetate diet containing 7.5% of GC seeds. These findings observed that GCs improved biochemical indicators of bones.

Bone loss is one of the most common adverse effects of glucocorticoid usage, even at low dosages. The major impact of glucocorticoids on bone is to suppress osteoblast activity, which results in a reduction in bone formation **(De Nijs, 2008).** Regarding the mean values of both bone mineral density (BMD) and bone mineral concentration (BMC), it could be noticed that employing prednisone acetate to induce osteoporosis led to a significant decline in BMD and BMC (g/cm^2) in the positive control group, as compared to the negative control group (0.09±0.02 and 0.16±0.03 vs.0.17±0.01 and 0.31±0.02 g/cm^2, respectively). The obtained results indicated that BMD and BMC increased gradually with the increasing levels of GC seeds. The best results of BMD and BMC of all tested groups were recorded for the group treated with 7.5% followed by 5% GC seeds. The results showed the positive effect of GC seeds in improving BMD and BMC due to their minerals content which stimulates the formation of bone osteoblasts and prevents bone resorption. Also, the Histopathological examination confirmed these results.

These results are in line with those reported by **Bukhari *et al.* (2018)** who indicated that *L. sativum* improves bone structure and reduces symptoms that occur with arthritis and body systems inflammation; where it was observed significant changes in BMD and improved T-Score at baseline after treatment with *L. sativum*. **Al-Tamimi *et al.* (2020)** mentioned that *Lepidium sativum* seeds elevated the serum calcium level. According to **Mabrouk *et al.* (2020)** GC seeds oil increased mineral and vitamin D concentrations, and improved histological structure and bone thickness in rats suffering from osteoporosis. **Alharbi *et al.* (2021)** demonstrated that *L. sativum* seeds improve bone healing and could be used as a supplemental or alternative therapeutic osteogenic agent in the therapy of bone fractures.

Table (3): Effect of GCs on serum calcium and phosphorus, femur bone calcium and phosphorus, BMD and BMC of female osteoporotic rats

Parameters / Groups	Serum Ca	Serum P	Femur bone Ca	Femur bone P	BMD	BMC
	mmol/L		mg/kg		g/cm^2	
Control -	5.04^{a} ±0.31	3.12^{a} ±0.37	50.2^{a} ±2.92	94.64^{a} ±2.50	0.17^{a} ±0.01	0.31^{a} ±0.02
Control +	2.19^{d} ±0.21	1.42^{d} ±0.15	30.62^{c} ±3.03	61.36^{d} ±4.66	0.09^{d} ±0.02	0.16^{d} ±0.03
2.5% GCs	3.13^{c} ±0.51	2.00^{c} ±0.50	31.15^{c} ±2.55	63.45^{d} ±5.32	0.13^{c} ±0.02	0.20^{c} ±0.02
5% GCs	4.12^{b} ±0.28	2.29^{bc} ±0.37	40.72^{b} ±2.19	72.33^{c} ±7.28	0.14^{bc} ±0.01	0.25^{b} ±0.01
7.5% GCs	4.64^{ab} ±0.41	2.86^{ab} ±0.61	46.65^{a} ±3.27	83.01^{b} ±7.11	0.16^{ab} ±0.01	0.27^{b} ±0.02

GCs: Garden cress seeds, BMD: Bone Mineral Density, BMC: Bone Mineral Concentration.
Means in the same column with different superscript letters are significantly different at p≤0.05

Kidney function

According to **Jeon (2008),** the kidney regulates calcium excretion by reabsorbing approximately 95% of filtered calcium through the renal tubules. Data presented in Table (4) showed the effect of GC seeds on serum uric acid, urea nitrogen, and creatinine of female rats suffering from osteoporosis. The mean values ± SD of uric acid, urea nitrogen, and creatinine (mg/dl) of the osteoporotic group (C+) were significantly (P≤0.05) increased as compared with the intact group (2.51±0.15, 60.31±4.54 and 1.55±0.09 vs. 1.57±0.07, 30.42±1.55 and 0.52±0.04 mg/dl, respectively). Treating osteoporotic groups with a prednisone acetate diet supplemented with 5% and 7.5% GCs decreased the levels of serum uric acid, urea nitrogen, and creatinine of rats suffering from osteoporosis, while the osteoporotic group of rats which treated with the low level of GCs didn't differ significantly in serum urea nitrogen and creatinine as compared with the positive control group. The results showed that the biologically active phytochemicals ingredients in garden cress could improve kidney functions in osteoporotic rats.

The present study is in harmony with **Al Hamedan (2010)** who found that oral administration of garden cress seeds extract and powder showed significantly lower values for serum urea and creatinine in rats. Similarly, **Nilesh *et al.* (2010)** discovered that administering *L.sativum* extract reduced serum creatinine and urea levels substantially. **Sakran *et al.* (2014)** reported that glucosinolates and other compounds in *L. sativum* seed are responsible for protecting cells of the liver and kidney against various toxins. Also, **Halaby *et al.* (2015)** mentioned that the diets fortified at 5% and 10% seeds powder of *L. sativum* helped to improve kidney and liver function compared with positive control groups.

Table (4): Effect of GCs on serum kidney functions of female osteoporosis rats

Parameters / Groups	Uric acid	Urea nitrogen	Creatinine
	mg/dl		
Control -	1.57 ± 0.07^{e}	$30.4+2\pm1.55^{d}$	0.52 ± 0.04^{d}
Control +	2.51 ± 0.15^{a}	60.31 ± 4.54^{a}	1.55 ± 0.09^{a}
2.5% GCs	2.32 ± 0.20^{b}	56.98 ± 4.66^{a}	1.49 ± 0.05^{a}
5% GCs	1.99 ± 0.12^{c}	49.82 ± 3.09^{b}	1.17 ± 0.04^{b}
7.5% GCs	1.79 ± 0.08^{d}	43.68 ± 3.23^{c}	0.89 ± 0.05^{c}

GCs: Garden cress seeds
Means in the same column with different superscript letters are significantly different at $p\leq0.05$.

Liver enzymes activity

Results illustrated in Table (5) revealed the effect of GC seeds on serum liver functions of osteoporotic female rats. The mean values ± SD of serum AST, ALT, and ALP **(u/L)** significantly increased

(P≤0.05) in the positive control group as compared to the negative control group (133.47±3.14, 57.23±3.71 and 177.84±4.52 vs. 70.18±4.41, 26.61±2.77 and 81.37±2.21 u/L, respectively), because of nourishing prednisone acetate diet. Feeding osteoporotic groups on a prednisone acetate diet containing different levels of GCs showed a significant decrease in the mean values of serum AST, ALT and ALP, as compared to the positive control group. The best result of liver enzymes activity was recorded for the group treated with 7.5% GC seeds.

These results were in line with those found by **Al-Asmari *et al.* (2015)** who showed that *L. sativum* seeds ethanolic extract improves liver enzymes significantly. Similarly, **Raish *et al.* (2016)** found that supplementation with *L. sativum* seeds extract ameliorated liver enzymes activity and reduced hepatic injuries and structural damage by decreasing oxidative stress, inflammation, and apoptosis in the liver had.

Table (5): Effect of GCs on serum liver enzymes activity of female osteoporosis rats

Parameters / Groups	AST	ALT	ALP
	u/L		
Control -	70.18±4.41^{e}	26.61±2.77^{d}	81.37±2.21^{e}
Control +	133.47±3.14^{a}	57.23±3.71^{a}	177.84±4.52^{a}
2.5% GCs	119.53±4.34^{b}	53.09±6.37^{a}	163.88±3.76^{b}
5% GCs	107.77±4.57^{c}	46.31±2.08^{b}	153.59±3.07^{c}
7.5% GCs	98.08±4.42^{d}	38.31±4.19^{c}	146.73±3.59^{d}

GCs: Garden cress seeds
Means in the same column with different superscript letters are significantly different at p≤0.05.

Histological Examination

Fig. (1) showed the histopathological examination of the femur right bone. Microscopically, the bone of rats from C- group which fed on a basal diet revealed the normal histological structure of trabecular

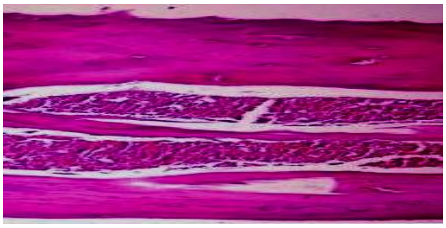
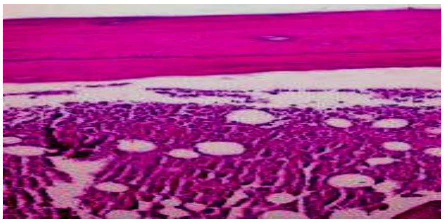
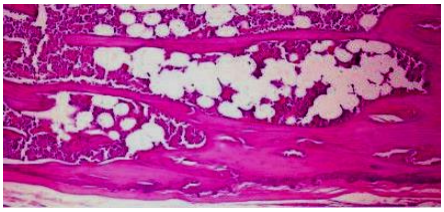
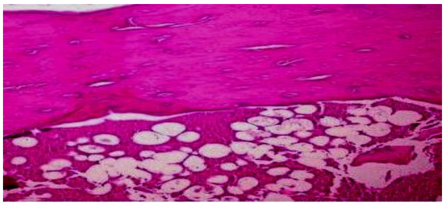
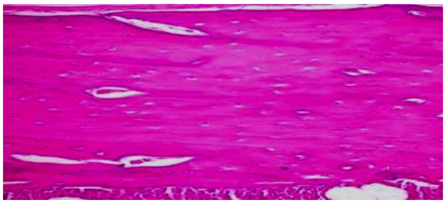
	**Photo (1): Control -** Normal histological structure of trabecular bone (H & E x 200).
	**Photo (2): Control+** Reduction in bone mass and the medullar cavity becomes enlarged (H & E x 200)
	**Photo (3): 2.5% GCS** Focal necrosis in bone cortex, few bone trabecular, and enlarged medullar cavity. (H & E x 200)
	**Photo (4): 5% GCS** Thick cortical bone and massive proliferation of osteoblasts, while some cavity still present. (H & E x 200)
	**Photo (5): 7.5% GCS** Obvious normal bone structure. (H & E x 200).

Fig (1): Histological examination of femur bone structure

bone (Photo1). While, the bone of rats suffering from osteoporotic C+ group showed a reduction in bone mass and the medullar cavity became enlarged (photo 2). In this respect, (Photos 3, 4 & 5) showed a gradual improvement of bone tissue, thick cortical bone and massive proliferation of osteoblasts in osteoporotic rats that received a prednisone acetate diet containing different ratios of GCs. The results agreed with **Mabrouk *et al.* (2020)** who found that consumption of GCs oil leads to enhancing the histological structure and bone thickness of osteoporotic rats.

Sensory properties of prepared peanut sweets with GCs

Peanut sweets are a popular product among all population groups in Egypt, especially when celebrating the Prophet's birthday. This kind of sweets can be made out of different nuts, the current study had chosen peanuts to make peanut sweets.

Sensory evaluation is a crucial indicator of potential consumer preferences. Table (6) presented the sensory characteristics of prepared peanut sweets with garden cress seeds. It could be observed that all formulations were acceptable in all sensory evaluation attributes (color, taste, flavor, texture, and general acceptability), there were no significant differences ($P \leq 0.05$) between the samples supplemented with 2.5% and 5% of GCs when compared with the control sample. While the flavor of the sample which contained 5% GCs slightly decreased as compared to the control peanut sweets sample. The addition of high levels of GCs by 7.5% caused significant changes ($P \leq 0.05$) in the same sensory evaluation attributes as compared to the control sample, but the sample was still acceptable. General acceptance score ranged from 8.24 to 6.78 by about 91.55 to 76.33%. It means that garden cress seeds can be used in the preparation of peanut sweets and any other product up to 7.5%.

These results are in harmony with **Umesha *et al.* (2015)** who indicated that biscuits prepared by supplementing garden cress oil at 5g/100 g by replacing fat were acceptable. **Alshehry (2019)** found that biscuits fortified with garden cress powder till 7.5 percent acceptance are the most popular among panelists.

Table (6): Sensory characteristics of prepared Sweet Peanuts with GCs

Sensory Characteristics	Control 0%	Sweet peanuts with GCs		
		2.5%	5%	7.5%
Color	8.53 [a] ±.262	8.47 [a] ±.371	8.41 [a] ±.200	8.08[b] ±.101
Taste	8.64 [a] ±.281	8.57 [ab] ±.301	8.31[ab] ±.383	7.93 [b] ±.380
Flavor	8.81 [a] ±.360	8.35 [ab] ±.281	7.68 [bc] ±.420	7.44 [c] ±.350
Texture	8.38 [a] ±.331	8.19 [a] ±.311	7.86 [ab] ±.321	7.12 [b] ±.510
General Acceptance	8.24 [a] ±.210	8.12 [a] ±.248	7.67 [ab] ±.385	6.87[c] ±.231

GCs: Garden cress seeds
Means with different letter within the same row are significantly different at P≤ 0.05

Conclusion

According to the findings, Garden Cress seeds are a good source of minerals and a variety of efficient prophylactic chemicals which significantly improved the osteoporosis bone markers. Garden Cress seeds may be utilized in many products as a functional food to reduce the risk of osteoporosis. Eventually, Garden Cress seeds can be applied up to 7.5% in functional foods.

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التأثيرات المحتملة لبذور حب الرشاد على عظام إناث الفئران المصابة بهشاشة العظام

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الملخص

ترتبط هشاشة العظام بارتفاع معدل انتشار الامراض والوفيات من جميع أنحاء العالم. أجريت هذه الدراسة لمعرفة التأثيرات المحتملة لبذور حب الرشاد على عظام إناث الفئران المصابة بهشاشة العظام. استخدمت الدراسة ثلاثون أنثى فأر من نوع الألبينو (200±10جم). قسمت الفئران إلى مجموعتين رئيسيتين، المجموعة الرئيسية الاولي (6 فئران) أصحاء تم تغذيتهم على الوجبة الاساسية كمجموعة ضابطة سالبة. تمت تغذية المجموعة الرئيسية الثانية (24 فأر) على غذاء اساسي يحتوي على 100مجم بريدنيزون أسيتات كمصدر للجلوكوكورتيكويد / كجم لمدة أسبوعين للحث على احداث هشاشة العظام. تم اختيار مجموعة واحدة منهم كمجموعة ضابطة موجبة، اما المجموعات (3 و4 و5) تم تغذيتها على وجبة بريدنيزون أسيتات تحتوي على مسحوق من بذور حب الرشاد بنسبة 2.5 و5 و7.5٪. أظهرت النتائج أن التركيب الكيميائي لمسحوق بذور حب الرشاد يحتوي على 2.75 و23.67 و22.81 و5.16 و8.42 و37.19 جم / 100 جم لكل من الرطوبة والبروتين والدهون والرماد والألياف الخام وإجمالي الكربوهيدرات على التوالي، بينما سجلت قيمة السعرات الحرارية 448.73 كيلو كالوري / 100 جم. وبلغ محتوى المعادن 309.21، 612.54، 341.10، 5.74 ملجم / 100 جم من الكالسيوم والفوسفور والمغنيسيوم والزنك على التوالي. أظهرت نتائج الدراسة البيولوجية تحسنًا معنويًا ($P \leq 0.05$) في قيم كل من الكالسيوم والفوسفور في مصل وعظم الفخذ في الفئران المصابة بهشاشة العظام والتي تغذت

على نظام غذائي أساسي يحتوي على نسب مختلفة من مسحوق بذور حب الرشاد، كما لوحظ زيادة كل من كثافة ومحتوى المعادن بالعظام مقارنة بالمجموعة الضابطة الموجبة. تحسنت وظائف الكلى والكبد معنوياً ($P \leq 0.05$) مقارنة بالمجموعة الضابطة الموجبة. أظهرت نتائج الفحص الهستولوجي للعظام تحسناً تدريجياً في أنسجتها. تم عمل حلوي الفول السوداني بالنسب 2.5 و5 و7.5٪ من مسحوق بذور حب الرشاد، وأشارت نتائج التقييم الحسي إلى أن جميع عينات حلوي الفول السوداني قد حظيت بقبول أكبر من 75٪. خلصت نتائج الدراسة الحالية الي أن مسحوق بذور حب الرشاد قد حسن من مضاعفات الاصابة بهشاشة العظام في الفئران، كما يمكن استخدامه بنسبة تصل إلى 7.5٪ كمكمل في النظام الغذائي لمرضى هشاشة العظام.

الكلمات المفتاحية: هشاشة العظام– حلوي الفول السوداني– بذور حب الرشاد –الكالسيوم– الفسفور– كثافة المعادن بالعظام.