



The effect of adding quinoa seeds powder to bread on the biochemical, nutritional and histological parameters on weaning rats

Rasha M. Arafa , Ghada M. Elseedy

Department of Home Economics, Faculty of Specific Education, Damietta University,
Damietta, Egypt

Abstract: Quinoa seeds can be very important for improving food supplies and as alternative food sources in several counties. The aim of this study was to investigate the effect of adding quinoa seeds powder (QSP) to bread on the biochemical, nutritional and histological parameters on weaning rats. Twenty four weaning male albino rats weighting an average of (160±10g) were divided into two main groups. The first main group (n=6) was fed on a diet consists of unsupplemented bread (white bread), as control group (C). The second main group was divided into three subgroups (6 rats per each); they were fed on a diet consists of bread supplemented with 5, 10 and 20g of QSP. Nutritional and biochemical Histological parameters indicated that supplemented bread with different levels of QSP by 5, 10 and 20% were accepted sensorial; the nutritive values of bread had gradually increased with increasing the replacement of QSP. Also, The results revealed that adding QSP to the bread improved weight gains, food efficiency ratio, serum glucose, kidney functions (uric acid, urea nitrogen and creatinine), lipid profile (cholesterol, triglycerides, HDL-c, LDL-c and VLDL-c), liver enzymes activities (AST; ALT and ALP) and selected minerals (calcium and total iron) especially, the group which were received bread supplemented with high levels from QSP. The results concluded that adding 20% of QSP to white bread has achieved the best results followed by a ratio of 10% when compared to the control group. Microscopically examined liver and kidney of rats fed on bread supplemented with 10 and 20% of QSP showed normal state in both hepatocytes and renal cells.

Key words: Functional food, serum glucose, kidney functions, liver enzymes activity, serum lipid profile, calcium, iron.

Introduction

The Food and Agriculture Organization FAO of the United Nations declared 2013 to be the international year of quinoa. Quinoa is a grain crop grown primarily for its edible seeds; the plant has originated in the Andean region of South America where it was domesticated 3,000-4,000 years ago for human consumption (**Tang *et al.*, 2015**). Because quinoa has a high nutritive value and desirable agricultural traits as tolerance to drought and salinity, it has gained increased demand in recent years.

The winter climate in Egypt favors good production of quinoa and it has already been cultivated in saline soils in Upper Egypt (**El-Assiuty *et al.*, 2014**). Quinoa is a species of the goosefoot genus (*Chenopodium quinoa*); it is one of the seeds considered as pseudocereals and even a pseudo-oilseed, this is also because of its unusual composition and exceptional balance between carbohydrates, fat and protein (**James, 2009**). Also, protein composed of 16 amino acids being rich in lysine, threonine, and methionine (**Aubrecht and Biacs 2001; Gorinstein *et al.*, 2002 and Wright *et al.*, 2002**). The protein is of high quality containing much higher content of lysine than cereals and even milk and being devoid of gluten makes it suitable for celiac patients (**Koziol, 1992; Repo-Carrasco and Serna 2011 and Stikic *et al.*, 2012**). Quinoa is moderately high in calories; starch present in the form of small granules; high viscosity make it useful for specialized industrial Applications (**Galwey *et al.*, 1990 and Tang *et al.*, 2002**). Moreover, quinoa rich in essential vitamins and minerals (**Konishi *et al.*, 2004**). Quinoa, according to sowing density, can grow from 1 to 3 m high. The seeds can germinate very fast; in a few hours after having been exposed to moisture. Quinoa crops is variety, it changes from white, yellow or light brown to red (**Valencia-Chamorro *et al.*, 2003**). Seed colors go from white to grey and black, potentially having tones of yellow, rose, red and purple and violet, often with very colorful mixes in the same panicles (**Vega-Gálvez *et al.*, 2010**).

Quinoa is an excellent example of 'functional food' that aims at lowering the risk of various diseases **Vega-Gálvez *et al.* (2010)**. On the other hand, (**Pa'sko *et al.*, 2009**) concluded that functional properties given by strongly active compounds including: flavonoids, phenolic acids, fat soluble vitamins, fatty acids, high dietary fiber, trace elements,

and other compounds which can prevent oxidative stress, particularly for all cell processes requiring antioxidant protection of membranes, like neuronal activity, with minerals and amino acid contents with potential implications for aiding memory and lowering anxiety under stressful conditions (**Gorinstein *et al.*, 2007**). Quinoa contains isoflavones (daidzein & genistein); these hormones could be recognized by human estrogen receptors and act as antagonists of vessel contraction and reduce arterial resistance; by that, quinoa offer an advantage over other plant foods for human nutrition (**James, 2009**).

According to **Ogungbenle (2003)**, the quinoa seed flour has good gelation property, water-absorption capacity, emulsion capacity and stability; as well contains high percentages of D-xylose and maltose, and low contents of glucose and fructose, which allows its use in malted drink formulations. Quinoa flour is commonly used in baby foods, soup, cookies, muffins, breakfast cereal, flakes, pasta, snacks, drinks, beer, diet supplements, and extrudates (**Caperuto *et al.*, 2000 and Dogan and Karwe, 2003**). Quinoa meat substitute has been introduced in Europe (**Launois, 2008**). Puffed grains of quinoa are produced commercially in Peru and Bolivia; in addition of that, the plant is sometimes grown as a green vegetable and its leaves are eaten fresh in combination dishes or cooked as rice; whole plant is also used a green fodder to feed cattle and poultry (**Repo-Carrasco and Serna, 2011**). The saponins obtained as a by-product in the processing of quinoa can be utilized by the cosmetics and pharmaceutical industries (**Bhathal *et al.*, 2015**). In general, quinoa seeds contain saponins in the seed coat; except sweet varieties, without saponin or containing less than 0.11% (**James, 2009**). Thus, the aim of this study was to investigate the effect of adding QSP to bread on the biochemical, nutritional and histological parameters of weaning rats.

Material and Methods

Materials

Quinoa Seeds, White flour (72% extract), yeast, salt and sugar were purchased from local market, A.R.C. Cairo, Egypt.

Casein, vitamins, minerals, cellulose and choline chloride were purchased from El-Gomhoerya Company for Drugs, Chemicals and Medical Instruments, Cairo, Egypt.

Twenty four weaning male albino rats (Sprague Dawley Strain) were obtained from Helwan farm, Ministry of Health and Population.

Methods

Preparation of white bread

White bread consists of wheat flour (90g), yeasts (5g), salt (2.5g), sugar (2.5g) and water.

Preparation of bread with QSP

White flour supplemented with QSP - after grinding seeds - (5, 10 and 20g /100 g) and used in the preparation of bread.

Sensory evaluation

Sensory evaluation was performed by invited ten panelists of staff members from Home Economics Department, Faculty of Specific Education, Damietta University, Damietta, Egypt. Each panelist was asked to evaluate unfortified and fortified bread samples with quinoa seeds, according to colour, odour, taste, texture and general appearance (**Abd El-Latif 1990**).

Chemical analysis

Moisture content, total protein, crude fat, fiber and ash were determined in both of quinoa seeds and bread samples according to the methods outlined in **A.O.A.C (1990)**. Carbohydrates (Nitrogen Free Extract NFE) content were calculated by difference, using the Equation: Carbohydrates (%) = [100 - (moisture + fat + protein + crude fiber + ash)]

Experimental design

Twenty four weaning male albino rats (60 ± 10 g) were kept in individual stainless steel cages under hygienic conditions and fed for one week on basal diet for adaptation in ad libitum. The basal diet in the preliminary experiment consists of 14% casein (protein > 80%), corn oil 4%, cellulose 5%, vitamin mixture 1%, salt mixture 3.5%, choline chloride 0.25% and the remainder is corn starch (**Reeves et al., 1993**). Vitamin mix and minerals mix prepared according to (**AOAC, 1975**).

After the period of adaptation on basal diet; the rats were divided into two main groups (n=6), the first main group was fed on a diet consists of un-supplemented bread (white bread), as control group (C). The second main group was divided into three subgroups (6 rats each); they were fed on a diet consists of bread supplemented with (5, 10 and

20g of quinoa seeds powder QSP). During the experimental period (40 days), the diets consumed and body weights were recorded.

At the end of the experiment, the animals were fasted overnight, then the rats were anaesthetized and sacrificed, and blood samples were collected from the aorta. The blood samples were centrifuged and serum was separated to estimate some biochemical parameters, i.e. uric acid (Fossati *et al.*, 1980), urea nitrogen (Patton and Crouch 1977), creatinine (Bartels and Bohmer, 1971), serum cholesterol (Allain *et al.*, 1974), triglycerids (Foster and Dumns, 1973), HDL-c (Lopes-Virella *et al.*, 1977), LDL-c and VLDL-c were calculated by the modified formula of Fried Wald *et al.*, 1972, serum glucose (Trinder, 1959) aspartate amino transferase (AST) and alanine amino transferase (ALT) (Reitman and Frankel, 1957) and alkaline phosphatase (ALP) (Kind and King, 1954), total iron (Ramsy, 1957) and calcium according to (Baginski, 1973).

Histological examination

Kidneys, heart, spleen and liver were removed from each rat, careful dissection, washed with saline solution, dried with filter paper and weighted to calculate organs to body weight %. The Kidneys and liver in each group was examined histopathologically, according to Sheehan and Hrapchak (1980).

Biostatistics

The obtained data was analyzed statistically for standard deviation and one way ANOVA test (Steel and Torri, 1980).

Results and Discussion

Data in Table (1) showed the sensory evaluation of unsupplemented and supplemented bread with QSP. Bread supplemented with 5, 10 and 20% of QSP resulted in non-significant changes in most sensory characteristics, as compared to unsupplemented bread (control). The color for the bread was significantly affected ($p < 0.05$) by the addition of different levels from QSP, whereas, the color of all bread samples showed non-significant changes between them. The flavor of all bread samples supplemented with 5, 10 and 20% had non-significant changes, as compared to control bread. On the other hand, bread supplemented

with 5% QSP recorded non-significant changes in taste, while other samples supplemented with 10% and 20% were slight lower than control bread. Results in the same table indicated that, Texture of bread samples decreased significantly when adding different levels of QSP to the bread. Statistical analysis showed non-significant changes in general acceptability between two bread samples supplemented with 5% and 10% of QSP as compared to the control, except the sample supplemented with the high level 20% of QSP; it had similar acceptance with other samples but non-significant as compared to the control. In general, results from total score occurred that, all samples obtained higher than 75%.

Table (1): Sensory evaluation of un-supplemented and supplemented bread with quinoa seeds powder

Samples	Sensory Characteristics					Total Score (100)
	Color (20)	Flavor (20)	Taste (20)	Texture (20)	General Acceptance (20)	
Bread control	19.166 ± 0.652 ^a	18.833 ± 0.983 ^a	19.500 ± 0.547 ^a	19.166 ± 0.752 ^a	19.500 ± 0.547 ^a	96.165 ± 0.983 ^a
Bread + 5% QSP	17.666 ± 1.032 ^b	18.000 ± 1.414 ^a	19.00 ± 0.894 ^{ab}	18.00 ± 1.095 ^b	18.666 ± 0.816 ^{ab}	91.333 ± 4.501 ^b
Bread +10% QSP	17.333 ± 0.816 ^b	17.833 ± 1.471 ^a	17.00 ± 1.264 ^{bc}	17.666 ± 0.516 ^b	18.833 ± 1.169 ^{ab}	88.665 ± 3.559 ^b
Bread +20% QSP	16.666 ± 0.816 ^b	17.500 ± 1.870 ^a	16.166 ± 0.752 ^c	16.333 ± 0.816 ^c	17.333 ± 0.816 ^{bc}	84.000 ± 3.521 ^c

QSP analyzed for its content and illustrated in Table (2). The percent content of moisture, total protein, fat, ash, crude fiber and carbohydrates of QSP were 8.74, 13.32, 5.64, 1.87, 3.34 and 67.09 respectively. Regarding quinoa's total energy; it is comparable 372.4 kcal/100g as shown in the same table.

Chemical composition and nutritive values of bread supplemented with QSP are presented in Table (3). From the obtained results, control sample containing 12.25% total Protein, 0.98% crude Fat, 0.91% crude fiber, 0.9% ash, 77.06% carbohydrates and 366 Kcal/100g total energy. From the same table, it could be observed that, addition of QSP with 5, 10 and 20% to the bread leading to improve the nutritive values. There was an increase in total protein content by about 16.3%, 21.6 and 26.5% for

QSP bread, compared to the control. Crude fat, fiber and ash content for the bread supplemented with QSP showed a percentage increase from 27.5, 15.4 and 11.1% to 94.8, 95.6 and 100% respectively. Whereas, the replacement of wheat flour with 5, 10 and 20% QSP caused gradually slightly decreased of carbohydrates and total energy values as compared to the control group.

Table (2): Gross chemical composition (g/100g) and nutritive values (Kcal/100g) of QSP.

Components	Moisture	Total Protein	Fat	Crude Fiber	Ash	Carb.	Total energy
QSP	8.74	13.32	5.64	1.87	3.34	67.09	372

Table (3): The effect of adding QSP on chemical composition and nutritive values of bread.

Component	Bread value (Control)	Bread + QSP (%)					
		5		10		20	
		value	% of change	value	% of change	value	% of change
Moisture (%)	7.9	8.2	3.8	8.2	3.8	8.4	6.3
Total Protein (%)	12.25	14.25	16.3	14.9	21.6	15.5	26.5
Crude Fat (%)	0.98	1.25	27.5	1.62	65.3	1.91	94.8
Crude Fiber (%)	0.91	1.05	15.4	1.14	25.3	1.78	95.6
Ash (%)	0.9	1.0	11.1	1.5	66.6	1.8	100
Carbohydrates (%)	77.06	74.25	- 3.6	72.64	- 5.7	70.61	- 8.5
Total Energy (Kcal/100g)	366.06	365.3	- 0.21	364.8	- 0.34	361.3	- 1.29

The Table (4) summarize the effect of adding quinoa seeds powder to bread on feed intake, body weight gain%, food efficiency ratio and organs weight / body weight % of weaning rats. Results showed non-significant changes in feed intake (g/day/rat) in all group of weaning rats fed on bread supplemented with QSP as compared to the control. From the obtained results in the same table (4), the mean values of BWG% for weaning rats fed on bread supplemented with 10% and 20% QSP recorded a significant increase ($p < 0.05$), While group of rats receiving a diet containing bread+5% QSP showed non-significant changes,

compared to control group. On the other hand, food efficiency ratio (FER%) changed by feeding on bread supplemented with 10% and 20% QSP, statistical analysis showed a significant increase in FER% compared with control group. Furthermore, results showed that, weights of kidney and liver showed non-significant difference in kidney and liver weight/body weight % for all weaning rats fed on bread supplemented with different levels of QSP, as compared to control group. Whereas, liver weigh/body weight % of weaning rats fed on bread + 20% QSP recorded a significant increase, as compared to control group.

Table (4): Effect of adding QSP to bread on feed intake, body weight gain%, food efficiency ratio and organs weight / body weight % of weaning rats.

Parameters Groups	Feed intake (g/day/rat)	FER (%)	BWG (%)	Organs weight / body weight (%)	
				Liver	Kidney
Bread control	12.159 ± 0.663 ^a	77.404 ± 7.582 ^b	0.113 ± 0.020 ^b	2.667 ± 0.269 ^b	0.693 ± 0.066 ^a
Bread +5% QSP	11.825 ± 0.435 ^a	80.529 ± 9.125 ^b	0.118 ± 0.017 ^b	2.527 ± 0.153 ^{bc}	0.686 ± 0.078 ^a
Bread +10% QSP	12.026 ± 0.472 ^a	124.051 ± 13.100 ^a	0.181 ± 0.004 ^a	2.229 ± 0.111 ^c	0.639 ± 0.029 ^a
Bread +20% QSP	12.230 ± 0.477 ^a	134.319 ± 3.022 ^a	0.197 ± 0.019 ^a	3.315 ± 0.121 ^a	0.667 ± 0.103 ^a

Values are expressed as mean ± SD. Values which have different letters in the same column differ significantly at p<0.05

The results in Table (5) Figure (1) revealed that, serum glucose (mg/dl) of all groups fed on bread supplemented with 5, 10 and 20% QSP recorded a significant decrease p<0.05, as compared to control group. The ratios of decreasing glucose percent were about 36.4%, 41% and 44.7%, respectively. Results in the same table and Fig. showed the effect of adding QSP to bread on serum Aspartate Amino Transferase (AST), Alanine Amino Transferase (ALT) and alkaline phosphatase (ALK). Supplemented bread with QSP by 5, 10 and 20% led to decrease the values of serum AST, ALT and ALP (u/l) gradually with increasing the level of addition. Ratios of decreasing AST, ALT & ALP percent were about 43.16%, 60% and 39.18% respectively, when weaning rats fed on bread supplemented with 20% QSP compared with bread control group.

Table (5): Effect of adding QSP to bread on serum glucose and liver enzymes activity of weaning rats

Groups	Glucose (mg/dl)	AST (U/l)	ALT (U/l)	ALP (U/l)
Bread control	134.750 ± 3.593 ^a	150.00 ± 7.615 ^a	45.00 ± 1.154 ^a	411.500 ± 10.344 ^a
Bread +5% QSP	85.750 ± 5.188 ^b	115.00 ± 5.715 ^b	34.250 ± 4.349 ^b	355.00 ± 30.670 ^b
Bread +10% QSP	79.500 ± 3.785 ^c	110.250 ± 5.619 ^b	20.250 ± 1.707 ^c	287.500 ± 10.115 ^c
Bread +20% QSP	74.500 ± 3.415 ^c	85.250 ± 2.217 ^c	18.00 ± 2.828 ^c	250.250 ± 7.762 ^d

Values are expressed as mean ± SD , Values which have different letters in the same column differ significantly at p<0.05

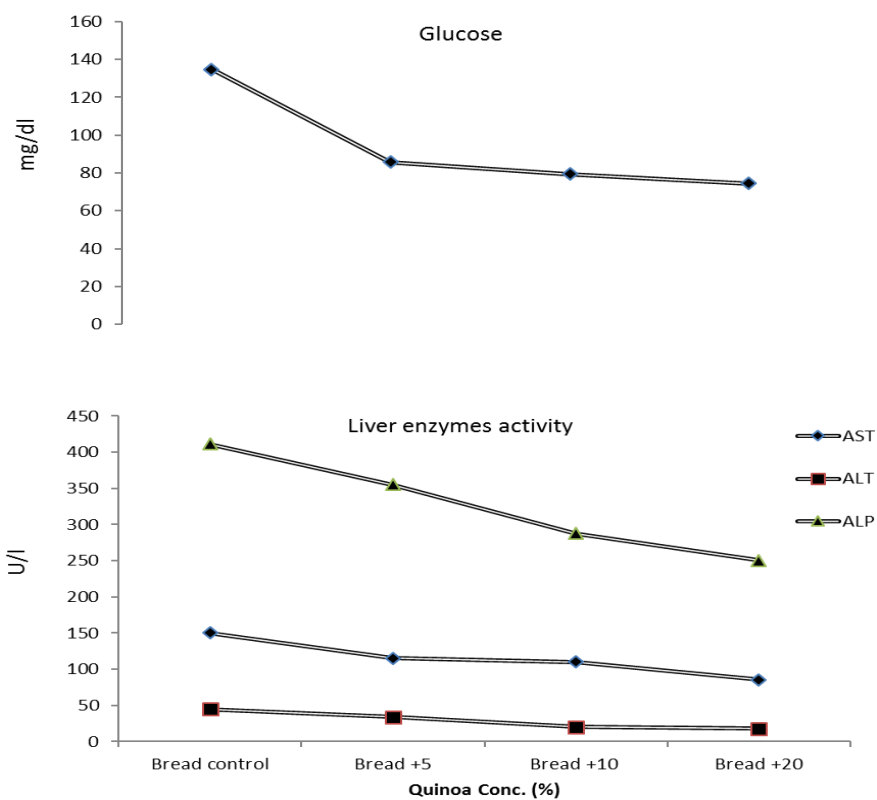


Figure (1): Effect of adding quinoa seeds powder to bread on serum glucose and liver enzymes activity

Data presented in Table (6) and Figure (2) the effect of adding quinoa seeds powder to bread on serum uric acid, urea nitrogen and creatinine o weaning rats. All groups which were fed on bread supplemented with QSP 5, 10 and 20% showed significant reduction in the mean values of uric acid and urea nitrogen at ($p < 0.05$), compared with control group. On the other hand, the lowest mean value of serum uric acid and urea nitrogen concentration were (1.650 ± 0.12^c and 26.750 ± 1.892^c) obtained from feeding rats with 20% QSP, as compared to control group and recorded the best results. Feeding rats on bread supplemented with 5% and 10% of QSP showed non-significant changes in creatinine, while group of rats which fed on bread + 20% QSP decreased significantly by about 19.05%, compared to control group.

Table (6): Effect of adding QSP to bread on kidney function of weaning rats.

Groups	Uric Acid (mg/dl)	Urea Nitrogen (mg/dl)	Creatinine (mg/dl)
Bread control	3.025 $\pm 0.206^a$	58.250 $\pm 5.737^a$	0.625 $\pm 0.050^a$
Bread +5% QSP	2.125 $\pm 0.170^b$	40.500 $\pm 7.681^b$	0.650 $\pm 0.100^a$
Bread +10% QSP	2.150 $\pm 0.208^b$	39.500 $\pm 2.645^b$	0.650 $\pm 0.057^a$
Bread +20% QSP	1.650 $\pm 0.129^c$	26.750 $\pm 1.892^c$	0.525 $\pm 0.05^b$

Values are expressed as mean \pm SD. Values which have different letters in the same column differ significantly at $p < 0.05$

Table (7): Effect of adding QSP to bread on serum lipid profile of weaning rats

Groups	Cholesterol (mg/dl)	Triglycerides (mg/dl)	HDL-c (mg/dl)	LDL-c (mg/dl)	VLDL-c (mg/dl)
Bread control	91.750 $\pm 3.304^a$	92.00 $\pm 4.546^a$	35.00 $\pm 2.00^d$	38.350 $\pm 1.369^a$	18.400 $\pm 0.909^a$
Bread+5% QSP	96.250 $\pm 3.774^{ab}$	79.750 $\pm 6.601^b$	45.750 $\pm 2.629^c$	34.550 $\pm 2.100^b$	15.950 $\pm 1.320^b$
Bread+10% QSP	90.500 $\pm 3.109^b$	63.750 $\pm 3.500^c$	59.500 $\pm 2.516^a$	18.250 $\pm 1.491^c$	12.750 $\pm 0.700^c$
Bread +20% QSP	78.750 $\pm 2.872^c$	51.500 $\pm 2.380^d$	52.00 $\pm 5.354^b$	16.450 $\pm 3.226^c$	10.300 $\pm 0.476^d$

Values are expressed as mean \pm SD. Values which have different letters in the same column differ significantly at $p < 0.05$.

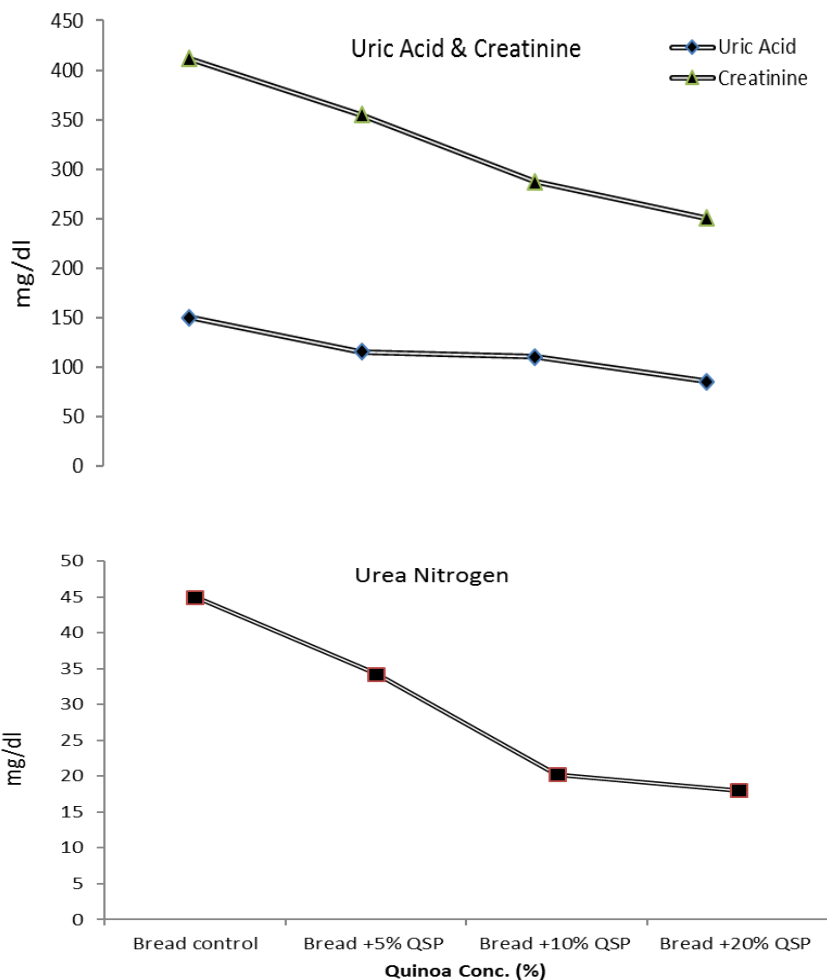


Figure (2): Effect of adding quinoa seeds powder to bread on kidney function

Finding in Table (8) Figure (4) presented the effect of adding quinoa seeds powder to bread on serum calcium and total iron of weaning rats. Addition of QSP to the bread at levels 5, 10 and 20% resulted in significant increase $p < 0.05$ in the mean values of serum calcium (mg/dl) and total iron (Ug/dl) as compared to control group. The best results of selected serum minerals were noticed in group of rats fed on bread supplemented with a highest level of QSP 20% by about 25.6% for serum calcium and 64.8% for total iron.

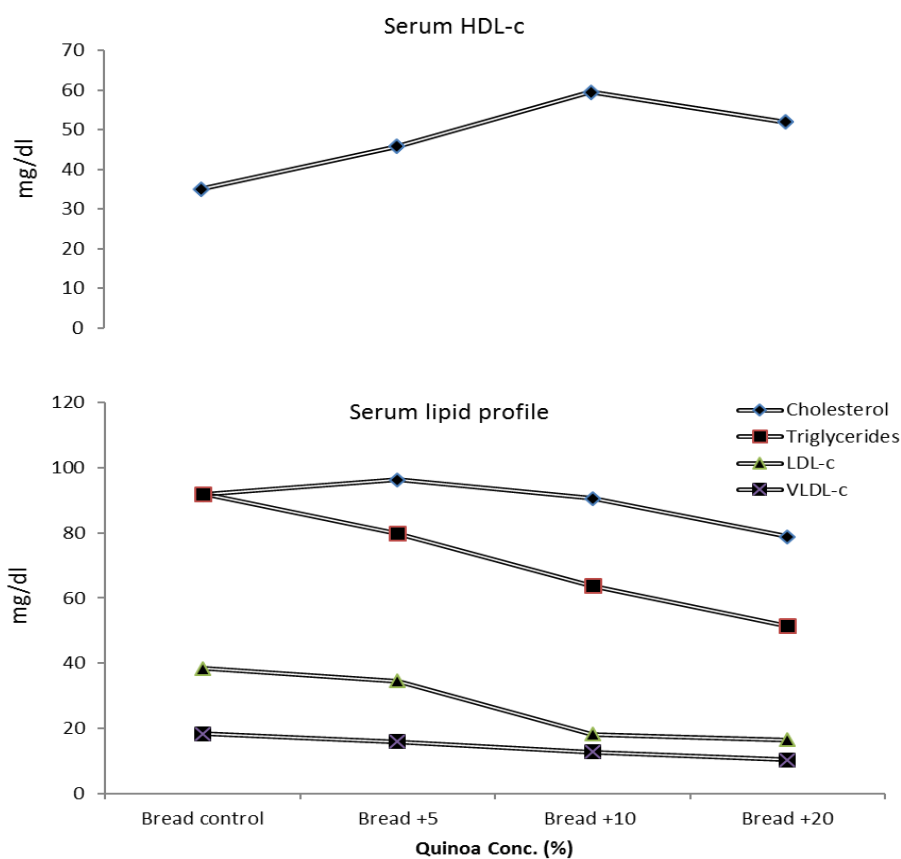


Figure (3): Effect of adding quinoa seeds powder to bread on kidney function

Table (8): Effect of adding QSP to bread on selected serum minerals of weaning rats

Groups	Calcium (mg/dl)	Total iron (Ug/dl)
Bread control	13.250 ± 3.340 ^{a b}	132.750 ± 4.716 ^d
Bread +5% QSP	8.975 ± 0.906 ^c	144.00 ± 8.205 ^c
Bread +10% QSP	9.875 ± 1.170 ^c	168.750 ± 6.849 ^b
Bread +20% QSP	10.550 ± 1.436 ^{b c}	218.750 ± 3.403 ^a

Values are expressed as mean ± SD. Values which have different letters in the same column differ significantly at p<0.05

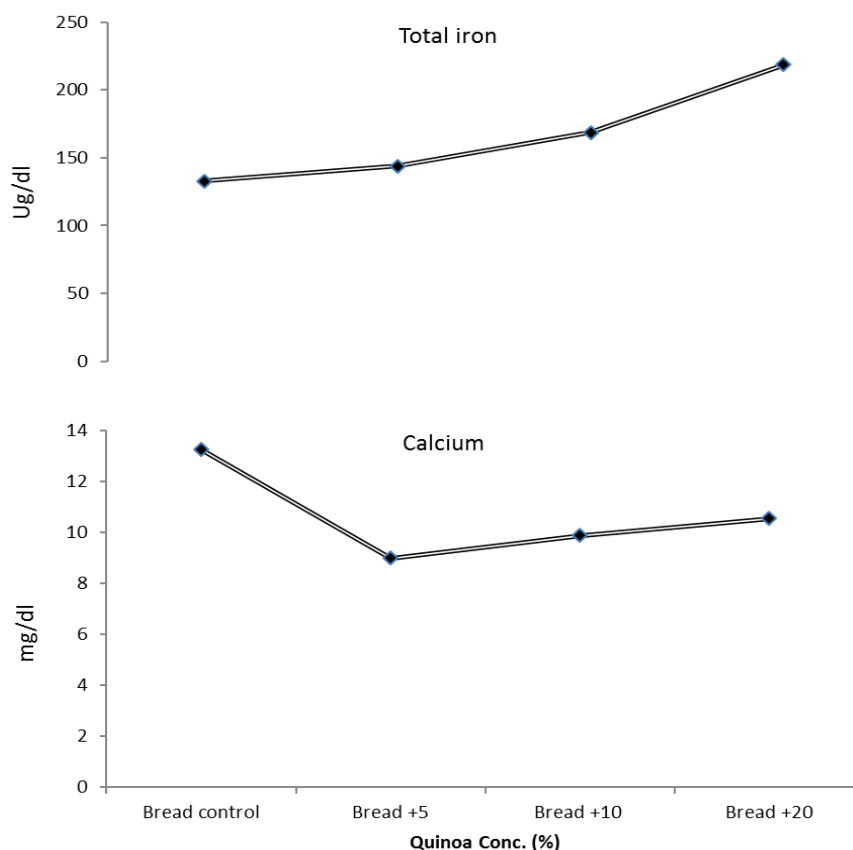


Figure (4): Effect of adding quinoa seeds powder to bread on serum calcium and total iron

Histopathological examination

Liver

Liver of rats from control group which fed on un-supplemented bread showed a slight congestion of hepatic sinusoids and cytoplasmic vacuolization of hepatocytes (Figures 5a). Some examined sections from group fed on bread supplemented with 5% QSP revealed cytoplasmic vacuolization of hepatocytes whereas, other sections showed hydropic degeneration of hepatocytes (Figures 5b). Liver of rats from group received bread + 10% QSP showed no histopathological changes, except

slight activation of Kupffer cells (Figures. 5c). On the other hand, liver of rat from group fed on bread supplemented with 20% QSP revealed the normal histological structure of hepatic lobule (Figures. 5d).

Kidney

Microscopically, kidneys of rat from control group which fed on un-supplemented bread revealed the normal histological structure of renal parenchyma (Fig. 6a). Some examined sections from group fed on bread supplemented with 5% QSP revealed no histopathological changes (Fig. 6b). Moreover, some examined sections from group of rats which received bread + 10% QSP as well as kidneys of rats from group fed on bread supplemented with 20% QSP revealed no histopathological changes (Figs 6c & 6d).

Discussion

Quinoa use could improve the intake of certain macromolecules and phytochemicals that are known to be beneficial to human health. It's become a staple in many health-conscious Americans' diets. According to Codex Alimentarius, quinoa is gluten-free products (gluten content < 20mg/kg) (**Zevallos et al., 2014**). Thus, they cannot be used alone for bread - making. However, they can be mixed with wheat flour in the preparation of bread with high nutritional value (**Morita, 2001**).

Consumption of seeds is the most common use of quinoa **James (2009)** mentioned that extruded corn grits–quinoa blends had high protein quality and solubility and an acceptable sensory evaluation. **Caperuto et al., (2000)** developed gluten-free quinoa spaghetti and the product was sensorially accepted by the panelists. Future uses can be wide-ranging, like textured and fermented products; there are many ways in which it can be consumed: cooked, as flour, extruded. Quinoa meat substitute has been introduced in Europe **James (2009)**. There are several developments with quinoa flour at a smaller scale, like bread, cookies, muffins, pasta, snacks, drinks, flakes, breakfast cereals, baby foods, beer, diet supplements, and extrudates (**Bhargava et al., 2006 and Dogan and Karwe, 2003**). The current study demonstrated that bread supplemented with different ratios of quinoa seeds powder by 5, 10 and 20% were accepted sensorial.

Figure 5: Liver of rats

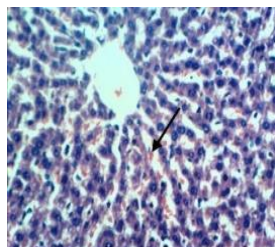


Fig. (5a)
A slight congestion of hepatic sinusoids and cytoplasmic vacuolization of hepatocytes (H & E X 400)

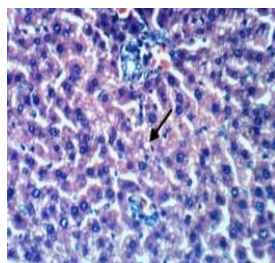


Fig. (5b)
Cytoplasmic vacuolization of hepatocytes whereas, other sections showed hydropic degeneration of hepatocytes

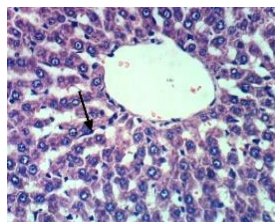
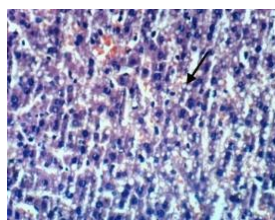


Fig. (5c)
No histopathological changes, except slight activation of Kupffer cells



(Figs. 5d)
Normal histological structure of hepatic lobule

Figure 6: Kidney of rats

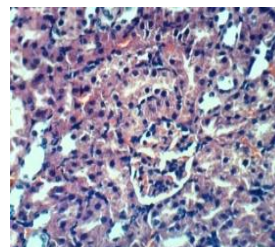


Fig. (6a)
Normal histological structure of renal parenchyma (H & E X 400).

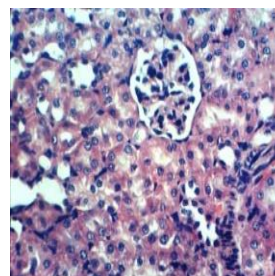


Fig. (6b)
No histopathological changes (H & E X 400).

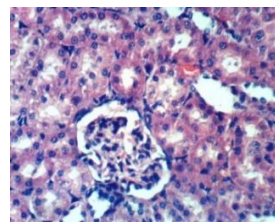


Fig. (6c)
No histopathological changes (H & E X 400).

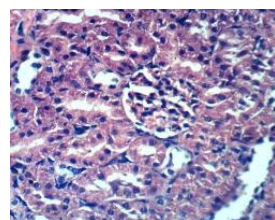


Fig. (6d)
No histopathological changes (H & E X 400).

The opportunity to supplement or completely replace common cereal grains (corn, rice and wheat) with a cereal of higher nutritional value (such as quinoa) is inherently beneficial to the public interest (**Brady *et al.*, 2007**). Protein content of four genotypes of quinoa grain ranges from 12.9% to 15.1%, with an average 15% (**Vega-G'alvez *et al.*,**

2010). Also, **Wright et al., (2002)** reported that a protein content of 14.8% and 15.7% for sweet and bitter quinoa, respectively, from Bolivia. On the other hand, quinoa protein can supply human with essential amino acids and the sulfur-containing amino acids. Lysine and methionine are found in concentrations that are unusually high compared to other cereals (**Bhargava et al., 2003**). According to studies of **Repo-Carrasco et al., (2003)** quinoa starch is the most important carbohydrate in all grains, making up approximately 58.1 to 64.2% of the dry matter. Also, its content of D-ribose and D-galactose and maltose would result in a low fructose glycemic index (**Vega-G'alvez et al., 2010**). Oil content in quinoa ranges from 1.8% to 9.5%, with an average of 5.0–7.2%, Linoleic acid is one of the most abundant polyunsaturated fatty acids identified in quinoa; polyunsaturated fatty acids have several positive effects on cardiovascular disease and improved insulin sensitivity (**James, 2009**). Studies reported that quinoa fat had a high content of oleic acid (24%) and linoleic acid (52%); all fatty acids present in quinoa are well protected by the presence of vitamin E, which acts as a natural antioxidant (**Ng et al., 2007**). Quinoa presents the highest percentage of fatty acids 50.2% for linoleic acid (ω -6) and 4.8% of linolenic acid (ω -3) (**Repo-Carrasco et al., 2003**). A lower ratio of omega-6: omega-3 fatty acids are more desirable for reducing the risk of cardiovascular disease, cancer, and inflammatory and autoimmune diseases **Tang et al., 2015**. Plus the neuronal activity of tryptophan amino acid and vitamin B complex can be powerful aids in brain function (**Vega-G'alvez et al., 2010**). In this investigation, the chemical composition and nutritive values of quinoa seeds powder were in the same line with these previous results.

Antioxidants are compounds that can delay or inhibit the oxidation of lipids or other molecules by inhibiting the initiation or propagation of oxidizing chain reactions. When added to foods, antioxidants minimize rancidity, delay the formation of toxic oxidation products, maintain nutritional quality and increase shelf life (**Vega-G'alvez et al., 2010**). Moreover, According to the USDA Nutrient Database, white bread is low in fiber which is an essential part of a healthy diet that lowers cholesterol level and keeps gastrointestinal tract working properly. The recommended daily intake of fiber is 14 grams for every 1,000 calories you consume while the average slice of white bread has 0.5 grams of fiber (**Solga, 2004**). Also, **Pasko and others (2010-a)** observed that groups fed with

diet supplemented with quinoa seeds showed significantly reduced serum total cholesterol (26%), LDL (57%), TG (11%) and glucose (10%), and the decrease in HDL levels was inhibited. Another study by **Pasko *et al.*, (2010-b)** showed the effect of diet supplemented with quinoa seeds on oxidative status in plasma and selected tissues of high fructose-fed rats study demonstrate that quinoa seeds can act as a moderate protective agent against potential of fructose-induced changes in rats by reducing lipid peroxidation and by enhancing the antioxidant capacity of blood (plasma) and heart, kidney, testis, lung and pancreas. **Farinazzi *et al.*, (2012)** studied the effects of quinoa on the biochemical and anthropometric profile and blood pressure in humans, parameters for measuring risk of cardiovascular diseases; twenty-two 18 to 45-year-old persons were treated daily for 30 days with quinoa in the form of a cereal bar. Results indicated that quinoa had beneficial effects since the levels of total cholesterol, triglycerides, and LDL showed reduction. It was concluded that the use of quinoa in diet can be considered beneficial in the prevention and treatment of risk factors related to cardiovascular diseases. These previous results were in agreement with the presented study which indicated that serum cholesterol, Triglycerides, LDL-C and VLDL-C declined gradually in weaning rats by adding different levels of quinoa seeds powder to the bread, at the same time, the level of serum HDL-C increased in comparison with control group.

White bread has a high glycemic index because it's made from refined grains that are rapidly absorbed during digestion, causing sharp spikes in blood sugar and insulin levels. A diet that includes a lot of white bread and other high-glycemic foods increases risk for weight gain, type 2 diabetes and heart disease. When blood sugar increased, it cause many trouble; a pro-inflammatory process that plays a role in a host of inflammatory diseases, cataracts and heart disease (**Queiroz *et al.*, 2012**). Carbohydrates from quinoa can be considered a nutraceutical food because they have beneficial hypoglycemic effects and induce lowering of free fatty acids. Studies made in individuals with celiac disease showed that the glycemic index of quinoa was slightly lower than that of gluten-free pasta and bread (**Berti *et al.*, 2004**). a low GI diet included quinoa; they observed an increase in HDL, gradual rises in blood sugar and insulin levels, so decrease type 2 diabetes risk factors. That result confirms the improving in serum glucose **Jenkins *et al.*, (2008)**. There is

some concern that diets higher in carbohydrates may be linked to the increased incidence of (nonalcoholic fatty liver disease NAFLD) (Alastair, 2013).

On the same direction, **Gordillo-Bastidas *et al.*, (2016)** reported that the general content of phytic acid in quinoa is low, it could act as an antioxidant: suppresses oxidant damage to the intestinal epithelium, prevents the formation of carcinogens and blocks the interaction of carcinogens with cells, controls cell division and increases the immune response by enhancing the activity of natural killer cells, inhibitor for renal stone development, cholesterol-lowering, affects the metabolic and detoxification capacity of the liver, reduces blood glucose and lipid, and hepatic lipid levels, inhibits calcification of cardiovascular system. In this aspect there is a confirmation of the current results with cited literature; adding quinoa seeds powder to the bread improved values of both liver enzymes and kidney function. **Also, Ricci *et al.*, (2011)** reported that a diet high in acidic foods, such as grains, forces the body to pull calcium from the bones. When researchers looked at how the diets of more than 500 women affected their bone density, they found that a diet high in refined grains was linked to bone loss. Wheat, in particular, is among the most potent sources of sulfuric acid, a powerful substance that quickly overcomes the neutralizing effects of alkaline bases. Vitamins are compounds essential for the health of humans. **Repo-Carrasco *et al.*, (2003)** showed that quinoa is rich in vitamin A, B2 and E. The content of vitamin E in quinoa is important since this vitamin acts as an antioxidant at the cell membrane level, protecting the fatty acids of the cell membranes against damage caused by free radicals. **Also, Vega-G'alvez *et al.*, (2010)** reported that quinoa has many vitamins, with 100 g of this grain containing: 0.4 mg of thiamine, 78.1 mg of folic acid, 1.4 mg of vitamin C, 0.20 mg of vitamin B6, and 0.61 mg of pantothenic acid. Additionally, many minerals in quinoa are found at concentrations greater than that reported for most grain crops; for example, iron, calcium, phosphorus, magnesium, iron, copper and zinc levels are higher than those of maize and barley (**Dini *et al.*, 2005 and Vega-G'alvez *et al.*, 2010**). The current study indicated that the supplementation of bread with different levels of quinoa seeds powder had increased serum total iron; led to enhancement level of hemoglobin; rise oxygen supply and reduce

anemic symptoms. Furthermore, the same study showed increased in serum calcium and superior the bone mineral density.

Conclusion

As a final point, quinoa could represent a supplementation crop that used to complement the diet in rural regions where energy-protein malnutrition affects a greater part of the population in certain developing countries. However, it is very important to increase and promote QSP Production, diversify production and enhance its consumption. Promoting quinoa consumption is to advise consumers of the good properties of quinoa and let them incorporate it in their daily diet as a healthy, nutritious, good tasting, and versatile food. It is necessary to be available on the market for the ordinary user, and scale them up to industrial level.

References

- AOAC (1975). Official Methods of Analysis of Assoc. of official agricultural chemists, 12th ed. Washington, DC.
- A.O.A.C. (1990): The Association of Official Analytical Chemists. 15th ed. Washington, DC.
- Abd El-latif, B.M. (1990): Improvement of some bakery products. [dissertation]. Zagazig, Faculty of Agriculture, Food Tech. Zagazig University, Egypt.
- Alastair, B. R.; Godin, J.P.; Minehira, K. and Kirwan, J. P. (2013): Increasing whole grain intake as part of prevention and treatment of nonalcoholic fatty liver disease. *Int J Endocrinol*.5: 1-13
- Allain, C. Z.; Poon, L. S. and Chan, C. S. (1974): Enzymatic determination of total serum cholesterol. *Clin. Chem.*, 20: 470-475.
- Aubrecht, E. and Biacs, P.A. (2001): Characterization of buckwheat grain proteins and its products. *Acta Aliment* 28:261-268
- Baginski, E.S. (1973): Method of calcium determination. *Clin .Chem. Acta*, 46:49.
- Bartels, H. and Bohmer, M. (1971). Creatinine standard and measurement of serum creatinine with picric acid. *Clin.Chem., Acta* 32: 81.
- Berti, C.; Riso, P.; Monti, L. and Porrini, M. (2004): In vitro starch digestibility and in vivo glucose response of gluten-free foods and their gluten counterparts. *Eur. J. Nutr.* 43(4): 198–204.

- Bhargava, A., Shukla, S., and Ohri, D. (2003): Genetic variability and heritability of selected traits during different cuttings of vegetable *Chenopodium*. *Ind. J. Genet. Pl. Breed* 63, 359–360.
- Bhargava, A.; Shukla, S.; and Ohri, D. (2006): *Chenopodium quinoa*—An Indian perspective. *Ind. Crops Prod.* 23: 73–87.
- Bhathal, S.; Grover, K. and Gill, N. (2015): Quinoa - a treasure trove of nutrients, *J Nut Res.*, 3(1): 45-49
- Bohmer, H.B.U.M. (1971). Micro- determination of creatinine. *Clin.Chem. Acta.*, 32:81-85.
- Brady, K.; Ho, C.; Rosen, R.; Sang, S. and Karwe, M. (2007): Effects of processing on the nutraceutical profile of quinoa. *Food Chem* 100:1209–1216
- Caperuto, L.; Amaya-Farfan, J. and Camargo, C. (2000): Performance of quinoa (*Chenopodium quinoa* Willd.) flour in the manufacture of gluten-free spaghetti. *Food Agri.* 8: 95–101.
- Dini, I.; Tenore, G.D. and Dini, A. (2005): Nutritional and antinutritional composition of *Kancolla* seeds: an interesting and underexploited andine food plant. *Food Chem* 92:125–132
- Dogan, H. and Karwe, M. (2003): Physicochemical properties of quinoa extrudates. *Food Sci. Techn. Int.* 9: 101–114.
- El-Assiuty, E.M.; Bekheet, F.M. And Fahmy, Z. M. (2014): First record of downy mildew of quinoa in Egypt. *Egypt. J. Agric. Res.*, 92 (3). 871.
- Farinazzi-Machado, F.M.V.; barbalho, S.M.; Oshiiwa, M.; Goulart, R. and Pessan Junior, O. (2012): Use of cereal bars with quinoa (*chenopodium quinoa* w.) to reduce risk factors related to cardiovascular diseases. *Food sci. technology* 32:239-244.
- Fossati, P.; Orenzi, L. and Berti, G. (1980): Egyptian colorimetric method of determination of uric acid in serum. *Clin. Chem.*, 26: 227.
- Foster, L. B. and Dumns, T. T. (1973): Determination of triglycerides. *J. Clin. Chem.*; 19:338 -353.
- Fried Wal, W.T.; Leve, R.I. and Fredrickson, D.S. (1972): Estimation of the concentration of low-density lipoprotein separation by three different methods . *Cli. Chem.*; 18: 499-502.
- Galwey, N.W.; Leakey, C.L.A.; Price, K.R. and Fenwick, G.R. (1990): Chemical composition and nutritional characteristics of quinoa (*Chenopodium quinoa* Willd.). *Food Sci Nutr* 42F:245–261
- Gordillo-Bastidas, E.; Díaz-Rizzolo, D.A.; Roura, E.; Massanés, T. and Gomis, R. (2016): Quinoa (*Chenopodium quinoa* Willd), from Nutritional Value

- to Potential Health Benefits: An Integrative Review. *J Nutr Food Sci* 6: 497-510.
- Gorinstein, S.; Pawelzik, E.; Gelgado-Licon, E.; Haruenkit, R.; Weisz, M. and Trakhtenberg, S. (2002): Characterization of pseudocereal and cereal proteins by protein and amino acid analysis. *J Sci Food Agric.*, 82:886-891
- Gorinstein, S.; Vargas, O.J.M.; Jaramillo, N.O.; Salas, I.A.; Ayala, A.M.; Arancibia-Avila, P.; Toledo, F.; Katrich, E. and Trakhtenberg, S. (2007): The total polyphenols and the antioxidant potentials of some selected cereals and pseudocereals. *Eur Food Res Technol.*, 225:321–328
- James, A.L.E. (2009): Quinoa (*Chenopodium quinoa* Willd.): composition, chemistry, nutritional, and functional properties. *Advances in Food and Nutrition Research*, (58): 1-32
- Jenkins, D.J.; Kendall, C.W.; McKeown-Eyssen, G.; Josse, R.G. and Silverberg, J. (2008): Effect of a low-glycemic index or a high-cereal fiber diet on type 2 diabetes: a randomized trial. *JAMA* 300: 2742-2753.
- Kind, P. R. N.; and King, E. J. (1954): Estimation of plasma phosphatase by determination of hydrolysed phenol with amino-antipyrine. *J. Clin. Path.*, 7: 322-326.
- Konishi, Y.; Hirano, S.; Tsuboi, H. and Wada, C. (2004): Distribution of minerals in quinoa (*Chenopodium quinoa* Willd) seeds. *Biosci Biotech Biochem* 68: 231-234
- Koziol, M. (1992): Chemical composition and nutritional evaluation of quinoa (*Chenopodium quinoa* Willd), *J. Food Comp. Anal.* 5: 35-68.
- Launois, A. (2008):
<http://www.bakeryandsnacks.com/news/ng.asp?id/485136>; Ancient grains on the rise.
- Lopes-Virella, M. F.; Stone, S.; Ellis, S. and Collwellm J. A. (1977): Cholesterol determination in high-density lipoproteins separated by three different methods. *Clin. Chem.*; 23 (5): 882-893.
- Morita, N. (2001): Quinoa flour as a new foodstuff for improving dough and bread *Journal of Applied Glycoscience*, v. 48, p. 263-270
- Ng, S.C.; Anderson, A.; Coker, J. and Ondrus, M. (2007): Characterization of lipid oxidation products in quinoa (*Chenopodium quinoa*). *Food Chem.*, 101:185-192
- Ogungbenle, H. (2003): Nutritional evaluation and functional properties of quinoa (*Chenopodium quinoa*) flour. *Int. J. Food Sci. Nutr.* 54: 153-158.

- Pa'ško, P.; Barto, N. H.; Zagrodzki, P.; Gorinstein, S.; Folta, M. and Zachwieja, Z. (2009): Anthocyanins, total polyphenols and antioxidant activity in amaranth and quinoa seeds and sprouts during their growth. *Food Chem* 115:994-998
- Pasko, P.; Zagrodzki, P.; Barton, H.; Chlopicka, J. and Gorinstein, S. (2010-a): Effect of quinoa seeds (*Chenopodium quinoa*) in diet on some biochemical parameters and essential elements in blood of high fructose-fed rats. *Plant Foods Hum Nutr.* 65: 333-338.
- Pasko, P.; Barton, H.; Zagrodzki, P.; Izewska, A.; Krosniak, M.; gawlik, M. and Gorinstein, S. (2010-b): Effect of diet supplemented with quinoa seeds on oxidative status in plasma and selected tissues of high fructose-fed rats. *Plant Foods Hum Nutr.* 65: 146-151.
- Patton, C. J. and Crouch, S. R. (1977): Enzymatic colorimetric method to determine urea in serum. *Anal. Chem.* 49:464.
- Queiroz, K.C.; Novato, S.I.; Cássia, G.A. (2012): Influence of the glycemic index and glycemic load of the diet in the glycemic control of diabetic children and teenagers. *Nutricion Hospitalaria.* 27(2): 510-515
- Ramsy, W.N.M. (1957): Determination of iron and iron binding capacity. *Clin. Chem. Act.* 2: 221.
- Reeves, P. G.; Nielsen, F. H. and Fahmy, G. C. (1993): Reported of the american institute of nutrition adhoc wriling committee on the reformulation of the ain-76 a rodent diet. *J. Nutr.* 123:1939-1951.
- Reitman, S. and Frankel, S. (1957): Determination of glutamate pyruvate transferase. *Am. J. Clin. Path.* 28: 56.
- Repo-Carrasco, R. and Serna, L.A. (2011): Quinoa (*Chenopodium quinoa*, Willd.) as a source of dietary fiber and other functional components *Cienc. Tecnol. Aliment., Campinas.* 31(1): 225-230
- Repo-Carrasco, R.; Espinoza, C. and Jacobsen, S.E. (2003): Nutritional value and use of the Andean crops quinoa (*Chenopodium quinoa*) *Food Rev Int* 19:179-189
- Ricci, G.; Canducci, E. and Pasini V. (2011): Nutrient intake in Italian obese patients: relationships with insulin resistance and markers of non-alcoholic fatty liver disease. *Nutrition* 27(6): 672-676.
- Sheehan, D. and Harpachak, B. (1980): Phory and bractec histotechnology. 2nd edn. Battle-Press; Ohio.
- Solga, S.; Alkhuraishe, A. R. and Clark J. M. (2004): Dietary composition and nonalcoholic fatty liver disease. *Digestive Diseases and Sciences.* 49(10): 1578-1583.

- Steel, R. G. and Torri, J. H. (1980). *Principal and procedures of statistical, biometrical approach*. Pbl. Mc Grew Hill Book Company. 2nd Ed. New York.
- Stikic, R.; Glamoclija, D.; Demin, M.; Vucelic-Radivic, B.; Jovanovic, Z. and Milokovic-Opsenica, D. (2012): Agronomical and nutritional evaluation of quinoa seeds (*Chenopodium quinoa* Willd.) as an ingredient in bread formulations. *J Cereal Sci.* 55:132-138
- Tang, H.; Watanabe, K. and Mitsunaga, T. (2002): Characterization of storage starches from quinoa, barley and adzuki seeds. *Carbohydr Polym* 49:13-22
- Tang, Y.; Li, X.; Zhang, B.; Chen, P.X.; Liu, R. (2015): Characterisation of phenolics, betanins and antioxidant activities in seeds of three *Chenopodium quinoa* Willd. genotypes. *Food Chem.* 166: 380-388.
- Trinder, P. (1959). Determination of blood glucose using U-Amino penzanone. *J. Clin. Path.* 22: 246-253.
- Valencia-Chamorro, S.A. (2003): Quinoa, in *encyclopedia of food science and nutrition*, ed. by Caballero B. Academic Press, Amsterdam, pp. 4895-4902.
- Vega-Gálvez, A.; Miranda, M.; Vergara, J.; Uribe, E.; Puente, L. and Martínez, EA. (2010): Nutrition facts and functional potential of quinoa (*Chenopodium quinoa* willd.), an ancient Andean grain: a review. *J Sci Food Agric.* 90(15):2541-2547.
- Wright, K.H.; Pike, O.A.; Fairbanks, D.J. and Huber, S.C. (2002): Composition of *Atriplex hortensis*, sweet and bitter *Chenopodium quinoa* seeds. *Food Chem Toxicol* 67:1383-1385
- Zevallos, V.F.; Herencia, L.I.; Chang, F.; Donnelly, S. and Ellis, H.J.; (2014): Gastrointestinal effects of eating quinoa (*Chenopodium quinoa* Willd.) in celiac patients. *Am J Gastroenterol* 109: 270-278.

تأثير اضافة مسحوق بذور الكينوا للخبز علي المقاييس الحيوية والغذائية والهستولوجية للفئران في مرحلة الفطام

رشا محمود عرفه ، غادة مسعد الصعيدي

قسم الاقتصاد المنزلي، كلية التربية النوعية، جامعة دمياط، دمياط ، مصر

ملخص البحث :

تعد الكينوا بذور ذات اهمية كبيرة في تحسين دعم الغذاء، كما تعد من مصادر الغذاء البديلة في عدة دول. اجريت الدراسة للتعرف علي تأثير اضافة مسحوق بذور الكينوا للخبز علي المقاييس الحيوية والغذائية والهستولوجية للفئران في مرحلة الفطام. استخدم في الدراسة ٢٤ فأر من ذكور الالبينو (160 ± 10 جم) في مرحلة الفطام، تم تقسيمها إلي مجموعتين رئيسيتين؛ المجموعة الرئيسية الأولى (٦) فئران تم تغذيتها علي غذاء اساسي يحتوي علي الخبز الابيض (مجموعة ضابطة)، والمجموعة الرئيسية الثانية (١٨) فأر تم تقسيمها الي ثلاث مجموعات فرعية؛ تم تغذيتها علي غذاء اساسي يحتوي علي الخبز الابيض مدعم بالنسب ٥ ، ١٠ ، ٢٠% من مسحوق بذور الكينوا. اشارت نتائج الدراسة أنه قد تم قبول الخبز الابيض المدعم بمسحوق بذور الكينوا حسيا، كما لوحظ ارتفاع القيمة الغذائية للخبز المدعم تدريجيا مع ارتفاع نسبة الاستبدال (الدعم). أظهرت النتائج أن إضافة مسحوق بذور الكينوا للخبز يحسن الوزن و نسبة كفاءة الغذاء ، كذلك ادي الي تحسن ملحوظ في وظائف الكلي (يوريا - حمض يوريك - كرياتينين)، مستوي الكوليسترول ودهنيات الدم ، انزيمات الكبد (الانين أمين ترانسفيريز ، اسبارتات أمين ترانسفيريز، الالكالين فوسفاتيز)، مستوي الجلوكوز ، وتحسن في مستويات الحديد والكالسيوم كما استنتجت الدراسة ان النسبة ٢٠% من بذور حبوب الكينوا المضافة للخبز الابيض قد حققت أفضل النتائج يليها النسبة ١٠% عند مقارنتها بالمجموعة الضابطة. كما اظهر الفحص المجهرى ان خلايا الكبد والكلي لدي الفئران التي تغذت علي الخبز الابيض المدعم بالنسب ١٠ ، ٢٠% طبيعية.

الكلمات المفتاحية: الاغذية الوظيفية - الجلوكوز - وظائف الكلي - انزيمات الكبد - دهنيات الدم - الكالسيوم - الحديد