Middle East Journal of Applied Sciences
ISSN 2077-4613
Volume : 05 | Issue : 02 | April-June | 2015
Pages: 573-586

Protective and Curative Effect of Garden Cress Seeds on Acute Renal Failure in Male Albino Rats

Mona S. Halaby, Mohammed H. Farag and Sara A. A. Mahmoud

Department of Nutrition and Food Science, Faculty of Home Economics, Helwan University, Cairo-Egypt

ABSTRACT

The main target of this study was to investigate the effect of two different concentrations (5% & 10%) of Garden Cress (GC) seeds powder on acute renal failure (ARF) of male albino rats. The experiment was carried out using forty nine male albino rats. These rats were put on ideal diet for one week before then divided into seven main groups including both curative and protective groups (seven rats of each). Curative groups: Group (1) Negative control group was fed on basal diet. Group (2) Positive control group was fed on basal diet + Cisplatin injection (5mg/kg body wt.) from the first day to induce ARF. Group (3) was fed as the group (2) + 5% (GC) seeds powder. Group (4) was fed as the group (2) + 10% (GC) seeds powder. Protective groups: Group (1) Negative control group was the same as Group (1) in curative group. Group (5) Positive control group was fed on basal diet + Cisplatin injection (5mg/kg body wt.) after 28 days. Group (6) was fed as the group (5) + 5% (GC) seeds powder. Group (7) was fed as the group (5) + 10% (GC) seeds powder.At the end of the experimental period rats were fasted overnight and sacrificed; blood samples were collected from the aorta to determine lipids profiles, also for kidney and liver functions. Besides, nutritional and biological parameters were recorded. Also, kidney was removed surgically for histopathological observation. From the obtained results we concluded that feeding ARF with GC seeds powder at 5% & 10% in curative groups and in protective groups improved the body weight gain, feed intake and feed efficiency ratio. Our results could be summarized that diet fortified at 5% and 10% GC seeds powder helped to improve blood lipid levels as well as reducing hazards on kidney and liver function compared with positive control groups (injected with Cisplatin) which were considered as a major risk factor for renal failure disease.Histopathological kidney observation proved that the last groups "curative & protective groups" fed on basal diet containing GC seeds powder at 10% GC showed proximal tubules suffering from mild individual cell necrosis and minimal interstitial inflammation.

Key words: Acute renal failure - Garden Cress seeds - Liver function - Kidney function - Histopathology.

Introduction

Kidney being one of the vital organs of human body performing the function of detoxification needs protection for healthy life. A large number of medicinal plants, natural products and dietary components have been evaluated as potential nephroprotective agents, however, very few herbs have been considered for nephroprotection it is often associated with a reduction in proteinuria (Yadav *et al.,* 2010 and Kasabe *et al.,* 2012).

Acute renal failure refers to the sudden and usually reversible loss of renal function, among the causes of acute renal failure, acute tubular necrosis, which occurs due to ischemia or nephrotoxins like Cisplatin is most common, accounting for 85% of the incidence. There is a continuous search for agents which provide nephroprotection against the renal impairment caused by drugs like Cisplatin for which allopathy offers no remedial measures showed by Zaveri *et al.,* (2011).

Cisplatin (Cis-diammine di-chloroplatinum II) is one of most potent anticancer drug, it is produced dose limiting nephrotoxicity and high dose of Cisplatin produce the impairment of kidney, causes decrease in renal blood flow, glomerular filtration rate and increases urea and creatinine level in blood. Kidneys have some antioxidant enzyme like superoxide dismutase (SOD), lipid peroxidase and glutathione (GSH), and catalase which protect kidney from free radicals like nitric oxide and superoxide etc. The Cisplatin is inhibited the activity of antioxidant enzyme in renal tissue like glutathione and SOD, and increase thiobarbuturic acid reactive substance (TBARS) showed by Ricardo *et al.,* (2005) and Yadav *et al.,* (2010).

The WHO estimates that 80% of people living in developing countries rely almost exclusively on traditional medicine for their primary health care needs. Medicinal plants form the back bone of traditional medicine and hence more than 3300 million people utilize medicinal plants on a regular basis. Demand for medicinal plants is increasing due to growing recognition of natural products being nontoxic, having no side effects. However, only a small proportion of hepatoprotective and nephroprotective plants as well as formulations used in traditional medicine are pharmacologically evaluated for their safety and efficacy revealed by Subramonium & Pushpangadan (1999) and Al-Sheddi *et al.,* (2013).

Corresponding Author: Mona S. Halaby, Soils, Department of Nutrition and Food Science, Faculty of Home Economics, Helwan University, Cairo-Egypt
E-mail: monahalaby03@yahoo.com

Garden Cress (*Lepidium Sativum*) is an annual herb, belonging to Brassicaceae family. It is a fast-growing, edible plant botanically related to watercress and mustard and sharing their peppery, tangy flavor and aroma. In some regions Garden Cress is known as garden pepper cress, pepper grass or pepperwort. It is also known as 'hab arachad' or "Thufa" (Manohar *et al.,* 2012 and Behrouzian *et al.,* 2014). Garden Cress is being cultivated as culinary vegetable in North America, Europe, and all over Asia including India, and in Egypt, Sudan and Saudi Arabia (Al-Sheddi *et al.,* 2013). The edible whole seed is known to have health promoting properties hence; it was assumed that these seeds can serve as raw material for functional foods (Snehal *et al.,* 2012; Rehman *et al.,* 2012 and Gaafar *et al.,* 2013). For this reason, the aim of the present investigation were to evaluate the biochemical, and biological changes that may occur in rats fed on basal diet with Cisplatin injection to induce acute renal failure, and to evaluate the potential effect of dietary supplementation with 5% or 10% (GC) seeds powder in adult male albino rats.

Materials and Methods

Materials:

Egyptian cultivar of *Garden Cress* seeds were purchased from Field Crops Research Institute, Ministry of Agriculture Giza, Egypt. Soybean oil and starch were purchased from the local market. Casein, cellulose, vitamins & minerals, dextrin, L-cysteine, choline chloride, and Cesplatin were obtained from the Cairo Company for Chemical Trading, Cairo, Egypt.

Forty nine male albino rats (Sprague Dawley strain) were obtained from the laboratory animal colony. Helwan, Cairo - Egypt. Weighting were approximately between (150-180g). Kits used to determine cholesterol, triglycerides, LDL-C, uric acid, urea nitrogen, creatinine, and transaminases produced by Egyptian American Company for laboratory service and supplied by Alkan Company. Histopathological examinations were obtained from National Research Center, Department of Pathology. Medical Research Division, Dokki, Cairo. Egypt.

Methods:

Preparation of dry Garden Cress seeds: was washed, dried and crushed using electric blender. These seeds powder were mixed with basal diet daily.

Chemical analysis of Garden Cress seeds:

Moisture, fat, protein, ash, crude fiber and tannins content were determined according to the method outlined in A.O.A.C. (2007). Total carbohydrates were determined by difference. Total fatty acids (Saturated and Unsaturated FA) were determined by "Hydrolytic Extraction Gas Chromatographic" according to the method described by ISO 5508 (1990) & ISO 5509 (2000).

Mineral contents including (Ca, P, Mg, Na, K, Cu, Fe, Mn & Zn) were determined according to the method described by Chapman & Pratt (1978). After complete digestion the minerals were determined using Unicam atomic absorption Spectrophotometer. Vitamins including (A, E, C, B_1, B_2, and Niacin) were assayed as recommended by J. Chrom. (1999 & 2006). Phenolic and Flavonoid compounds were determined by HPLC according to the methods of Goupy *et al.,* (1999) and Mattila *et al.,* (2000). Antioxidant activity and total carotenoid were determined according to the methods described by Politeo *et al.,* (2006) and Horwitz & Latimer (2007).

Experimental design:

Rats were adapted for one week prior to commencement of the experiment. Water was introduced ad-libitum. Rats were divided into seven groups including both curative and protective groups (seven rats of each).

Curative groups: Group (1) Negative control group was fed on basal diet according to Reeves *et al.,* (1993). Group (2) Positive control group was fed on basal diet + Cisplatin injection (5mg/kg body wt.) from the first day according to yogesh *et al.,* (2010). Groups (3 & 4) were fed as the group (2) + 5% & 10% (GC) seeds powder. Protective groups: Group (1) Negative control group was the same as Group (1) in curative group. Group (5) Positive control group was fed on basal diet + Cisplatin injection (5mg/kg body wt.) after 28 days. Groups (6 & 7) were fed as the group (5) + 5% & 10% (GC) seeds powder.

Feed intake, Body Weight Gain and Feed Efficiency Ratio were determined according to Chapman *et al.,* (1959).

Blood Sampling:

At the end of the experiment period, the rats were fasted overnight then the rats were anaesthetized and sacrificed and blood samples were collected from the aorta. The blood samples were centrifuged for 15 minutes at 3000 rpm to separate the serum. The serum was carefully separated into dry clean Wassermann tubes by using a Pasteur pipette and kept frozen at -20°C till analysis.

Biochemical analysis of serum:

Serum samples were used for the determination of total cholesterol (Allain *et al.,* 1974), triglycerides (Fassati & Prencipe 1982), HDL-C (Lopes, 1977), LDL-C and VLDL-C were calculated by using the method of Friedewald *et al.,* (1972). Uric acid, urea nitrogen and creatinine were determined according to the methods described by Fossati *et al.,* (1980), Patton & Crouch (1977), Bartels *et al.,* (1972), respectively. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined according to Retiman & Frankel (1957).

Histopathological Examination:

Tissues from kidney of the sacrificed rats were examined as described by Drury & Walligton, (1980).

Statistical analysis:

Results are expressed as mean ± SD. Data were statistically analyzed for variance using one-way analysis of variance "ANOVA" according to Armitage & Berry, (1987). Computer software system SPSS (version 15) was used for these calculations.

Results and Discussion

Chemical composition of raw materials:

Garden Cress seeds powder (GC) was investigated on dry weight basis. The following parameters in Table (1) were determined for moisture, fat, crude protein, ash, total carbohydrate and crude fiber, the ratios were 6.73; 32.28; 21.61; 4.83; 27.80 and 6.75 (g/ 100g DW), respectively. The present results are in agreement with those Gokavi *et al.,* (2004); Kadam *et al.,* (2012); Agarwal & Sharma (2013) and confirmed by Doke & Guha (2014) they published that the chemical composition of LS seeds indicates the presence of high amounts of protein (22.47%-25.00%) and lipids (14.00%-28.03%) which indicated that seeds have high food energy, with low moisture content (3.92%-4.14%) which an index of stability quality and increased shelf life of seeds, also, crude fiber (6.75%-16.50%), Ash (4.25%-4.65%), and carbohydrates (32.87%-54.00 %) respectively.

Results in the same table pointed out that GC seeds contained the high levels of total antioxidants activity and tannins, with the ratios 96.64±0.15 DPPH (μg/mL) and 13.95±0.21 (mg/100g), respectively. In fact, antioxidants are an important part of the defense system of the human body and help to cope with oxidative stress caused by reactive oxygen species. Our results are in agreement with Matthaus & Angelini (2005); Oz, (2011) and Bhasin et al. (2011) they confirmed that antioxidant activity of Garden Cress seeds was one of the supportive parameter in the nephron protective activity. It is well known that the chemical constituents, antioxidant capacity and phenolic content (tannins) of plants depend on several factors such as different genotype, growing condition, agronomic practices employed, season, maturity, post-harvest storage and processing conditions and solvent used for extraction. These factors may explain the differences found among our samples and those analyzed in previous studies Zia-Ul- Haq *et al.,* (2012) and Agarwal & Sharma (2013).

Table 1: Chemical composition of Garden Cress seeds powder (g / 100g dry weight basis)

Component (%)	Garden Cress seeds
Moisture	06.73 ± 0.05
Fat	32.28 ± 0.01
Crude protein	21.61 ± 0.088
Ash	04.83 ± 0.01
Total carbohydrates	27.80 ± 0.21
Crude fiber	06.75 ± 0.4
Total antioxidant activity DPPH (μg/mL)	96.64 ± 0.15
Tannins (mg/100g)	13.95 ± 0.21

The results given in Table (2) indicated that the nutritional activity in GC seeds is usually related to the particular elements, it contained good amounts of vitamins including (A, E, C, B_1, B_2, and Niacin), the values of vitamins with an average of 90.00 (μ/100g); 258.74 (ppm); 10.62; 00.59; 00.61 and 14.30 (mg/100g), respectively. Results in Table (2) are comparable to those of earlier workers for GC seeds (Yadav *et al.,* 2010) they confirmed that GC seeds contained good amounts of vitamins (β-carotene, riboflavin, niacin, and ascorbic acid). Moreover, Sharma & Agarwal (2011) published that total tocopherol content was 139.73 ± 0.91 mg/100g for L. Sativum, and it acts as biological scavengers of free radicals and could prevent diseases, besides possessing an important nutritional function for human beings as a source of vitamin E confirmed by Zia-Ul-Haq *et al.,* (2012).

Table 2: Vitamins composition of Garden Cress seeds powder (dry weight basis)

Component %	Garden Cress seeds
Vitamin A (µ/100g)	90.00
Vitamin E (ppm)	258.74
Vitamin C (mg/100g)	10.62
Thiamine (mg/100g)	00.59
Riboflavin (mg/100g)	00.61
Niacin (mg/100g)	14.30

Macro and micro nutrient elements can play an important role in many metabolic processes and functions throughout the life cycle. The data given in Table (3) illustrated that the GC seeds contained the various values of macro and micro-elements including calcium (Ca); Phosphorous (P); magnesium (Mg); sodium (Na); potassium (K); copper (Cu), iron (Fe), manganese (Mg) and zinc (Zn). Results indicated that GC contained the values of mineral with an average of 480.72; 637.25; 631.06; 36.25; 1635.62; 12.51; 28.82; 46.07 and 62.69 (mg/100g DW), respectively. These results were in agreement with those reported by Gokavi *et al.,* (2004); Yadav *et al.,* (2010) and Agarwal & Sharma (2013).

Table 3: Macro and Micro-elements of Garden Cress seeds powder (mg/100g dry weight basis)

Macro-elements	
Calcium (Ca)	480.72
Phosphorous	637.25
Magnesium (Mg)	631.06
Sodium (Na)	36.25
Potassium (K)	1635.62
Micro-elements	
Copper (Cu)	12.51
Iron (Fe)	28.82
Manganese (Mn)	46.07
Zinc (Zn)	62.69

Types and concentrations of fatty acids extracted from the *GC* seeds using gas chromatography (*GC*) give in Table (4). The results revealed that the total saturated fatty acid, monounsaturated and polyunsaturated fatty acids were 16.71; 42.61 and 40.68 (g/ 100g), and the most abundant poly and mono-unsaturated fatty acids were Linolenic (C18:3; 29.39%) and Oleic acids (C18:1; 24.65%), then followed by Gadoleic (C20:1; 12.27%) and Linoleic (C18:2; 11.29%) acids. Also, the results revealed that the most abundant saturated fatty acid in *GC* was Palmitic acid (C16:0; 8.36%). Results detected other quantities of fatty acids including Erucic (C22:1; 4.49%), Arachidic (C20:0; 3.61%) and Stearic (C18:0; 3.11%) acids were among the minor FA found in *GC* seeds. The obtained data were in the line with those of Moser *et al.,* (2009) and Gaafar *et al.,* (2013).

Table 4: Fatty acids composition of Garden Cress seeds powder

Component of fatty acids (g/100 g)		Garden Cress seeds
Saturated Fatty acid	Palmitic acid	8.36
	Margaric acid	0.03
	Lignoceric acid	0.70
	Stearic acid	3.11
	Arachidic acid	3.61
	Behenic acid	0.90
Mono-unsaturated Fatty acid	Oleic acid	24.65
	PalmitiOleic acid	0.34
	Erucic acid	4.49
	Gadoleic acid	12.27
	Nervonic acid	0.86
Poly-unsaturated Fatty acid	Linoleic acid	11.29
	Linolenic acid	29.39
Total saturated Fatty acid		16.71
Total Monounsaturated Fatty acid		42.61
Total polyunsaturated Fatty acid		40.68

Types of flavonoid and polyphenolic compounds in *GC* seeds give in Tables (5 & 6). It obvious that the abundant flavonoid compounds in *GC* seeds was Kampferol and Narengin which were at concentration of 709.66 and 610.55 (ppm). Querciterin; Apignin; Narenginin and Hispertin were the moderate abundant flavonoid compounds in *GC* seeds which were at concentration of 123.73, 96.34, 54.42, 44.45 (ppm), while, the lowest abundant were Rosmarinic; 7-Hydroxyflavon and Quercetin which were at concentration of 33.40; 21.19 and 13.99 (ppm), respectively. The results reported previously by Nayak *et al.,* (2009) and Ghante *et al.,* (2011) that flavonoids, cardiotonic glycosides, glucosinolates, sterols, tannins, and triterpene are important phytochemical constituents, which impart pharmacological characteristics to Garden Cress seed.

The results given in Tables (6) indicated that the Benzoic acid, Pyrogallol, Epicatechen, Protocatechuic acid, Salicylic acid, Catechol, Ellagic acid, Caffiene and Vanillic acid which were the abundant at concentrations of 186.94; 137.51; 135.97; 102.14; 91.50; 65.64; 58.77; 46.22 and 43.34 (ppm), respectively. While; Coumarin, Ferulic acid, P.OH. benzoic, Caffeic acid, Catechein and Chlorogenic were the moderate abundant polyphenolic compounds in GC seeds which were at concentration of 30.28, 25.84, 21.47, 20.45, 16.52 and 16.07 (ppm), also, the lowest abundant were Gallic acid and Syrengic acid which were at concentration of 3.24 and 2.54 (ppm), respectively. The obtained data were in the line with those of Indumathy & Aruna (2013) and Doke & Guha (2014) they published that the antioxidant compounds like phenolic acids, polyphenols and flavonoids scavenge free radicals such as peroxide, hydro peroxide or lipid peroxyl and thus inhibit the oxidative mechanisms that lead to degenerative diseases.

Table 5: Types of flavonoid compounds of Garden Cress seeds powder

Flavonoids (ppm)	**Garden Cress seeds**
Narengin	610.55
Rosmarinic	33.40
Querciterin	123.73
Narenginin	54.42
Quercetin	13.99
Hispertin	44.45
Kampferol	709.66
Apignin	96.34
7-Hydroxyflavon	21.19

Table 6: Types of polyphenolic compounds of Garden Cress seeds powder

Polyphenolic compounds (ppm)	Garden Cress seeds
Syrengic acid	2.54
Pyrogallol	137.51
Gallic acid	3.24
Protocatechuic acid	102.14
Catechol	65.64
Catechein	16.52
Chlorogenic	16.07
P.oH. benzoic	21.47
Epicatechen	135.97
Caffeic acid	20.45
Vanillic acid	43.34
Caffiene	46.22
Ferulic acid	25.84
Benzoic acid	186.94
Salicylic acid	91.50
Coumarin	30.28
Ellagic acid	58.77
Cinnamic	10.67

Biological evaluation

The mean values of rats body weight gain (BWG); their feed intake (FI) and feed efficiency ratio (FER) were summarized in Table (7). Data presented could be observed that there were significant decrease in BWG; FI and FER for control positive group (11.48±1.45; 9.79±0.14 and 0.059±0.06) after the single dose of Cisplatin (5mg/kg) as compared to the negative control group (19.7±2.04; 11.82±0.22 and 0.198±1.15), respectively. Weight loss with reduction of FI and FER during Cisplatin treatment (from the first day= curative group) may be due to gastrointestinal toxicity and by reduced ingestion of food confirmed by Mora *et al.,* (2003) and Yogesh *et al.,* (2011).

Feeding acute renal failure with GC seeds powder at 5% & 10% from the first day in curative groups showed significant increase values of BWG %; FI and FER compared to control positive group. Results revealed that the mean values and ± SD for curative group fed at 5% GC were 13.29±1.09; 10.07±0.23 & 0.098±0.16%. While, the mean values and ± SD for curative group fed at 10% GC were 15.26±2.06; 10.95±0.41 & 0.089±0.75% respectively, compared to the control positive group (11.48±1.45; 9.79±0.14 and 0.059±0.06), respectively.

Table 7: Effect of feeding acute renal failure rats on Garden Cress seeds on body weight gain %; feed intake and Feed efficiency ratio

Groups	Parameters	BWG (g)	FI (g/day)	(FER)
Control (-ve)		19.7^{a}± 2.04	11.82^{a}± 0.22	0.198^{a}± 1.15
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		11.48^{d}± 1.45	9.79^{c}±0.14	0.059^{d}± 0.06
Curative groups	Control (+ ve) + 5% GC	13.29^{c}± 1.09	10.07^{bc}±0.23	0.098^{b}± 0.16
	Control (+ ve) + 10% GC	15.26^{b}± 2.06	10.95^{b}±0.41	0.089^{c}± 0.75
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		16.64^{c}± 3.74	10.30^{c}±0.20	0.106^{b}± 0.25
Protective groups	Control (+ ve) + 5% GC	16.80^{bc}± 3.26	10.79^{bc}±0.15	0.038^{c}± 0.12
	Control (+ ve) + 10% GC	16.87^{b}± 3.21	10.83^{b}±0.21	0.033^{d}± 0.98

Moreover, results revealed that protective groups which fed on basal diet containing GC seeds powder at 5% & 10% then injected with Cisplatin after 28 days, improved the BWG %; FI and FER, compared with control positive group. The mean values for protective group fed on 5% GC were 16.80±3.26; 10.79±0.15 & 0.038±0.12; and the mean values for protective group fed on 10% GC were 16.87±3.21; 10.83±0.21 & 0.033±0.98 compared with control positive group 16.64±3.74; 10.30±0.20 & 0.106±0.25 respectively. These results are in harmony, with those obtained by the previous studies (Diwakar *et al.,* 2008 and Gaafar *et al.,* 2013) they reported that *Lepidium sativum* seeds increase weight gain as they are found to contain 18-24% of fat. Thirty four percent of the total fatty acids are alpha linolenic acid; which could give it nutritional advantages confirmed by Al- Hamedan, (2010) and Doke & Guha (2014).

Effect of feeding on Garden Cress seeds on lipid profile of acute renal failure rats:

Results in (Table 8) indicated that there were significant changes in the serum levels of Triglyceride (TG) and Total Cholesterol (TC) of negative control group when compared with ARF rats (positive control group), the values were 115.08±3.61 & 145.71±5.35 vs 180.71±4.16 & 239.28±4.01 (curative group) and the values were 115.08±3.61 & 145.71±5.35 vs 148.57±0.29 & 206.56±3.61 (protective group) respectively.

Feeding curative groups with 5% and 10% of GC induced significant decrease ($p < 0.05$) in triglyceride and total cholesterol (173.57±3.04 & 168.57±3.39 (5% GC) & 156.42±5.12 & 165.71±5.04 (10% GC) compared with those of the control positive group 180.71±4.16 & 239.28±4.01 respectively. Moreover, with protective groups, the values were 139.16±0.41 & 182.14±2.07 (5% GC) and 124.28±0.35 & 177.00±3.12 (10% GC), compared with those of the control positive group 148.57±0.29 & 206.56±3.61 respectively. In fact, rats which fed on GC seeds powder at 5% & 10% had lower mean values of TC and TG compared with the positive control group.

Confirmed in the previous studies that the Cisplatin injection rats caused acute renal failure, and concluded that a significant increase in serum cholesterol and triglycerides (Al-Hamedan, 2010), confirmed by Diwakara, *et al.,* (2010) and Chauhan *et al.,* 2012) that GC seeds supplementation in the human diet is efficient in depressing cholesterol and triglycerides.

Table 8: Effect of feeding acute renal failure rats on garden cress at different ratios on Triglyceride and Total Cholesterol

Groups	Parameters	Triglyceride mg/dl	Cholesterol mg/dl
Control (-ve)		115.08^{d} ± 3.61	145.71^{d} ± 5.35
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		180.71^{a} ± 4.16	239.28^{a} ± 4.01
Curative groups	Control (+ ve) + 5% GC	173.57^{b} ± 3.04	168.57^{b} ± 3.39
	Control (+ ve) + 10% GC	156.42^{c} ± 5.12	165.71^{c} ± 5.04
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		148.57^{a} ± 0.29	206.56^{a} ± 3.61
Protective groups	Control (+ ve) + 5% GC	139.16^{b} ± 0.41	182.14^{b} ± 2.07
	Control (+ ve) + 10% GC	124.28^{cd} ± 0.35	177.00^{c} ±3.12

**Results are expressed as means ±SD.*
**Values at the same column with different letters are significant at (p<0.05).*

Results in Table (9) indicated that there were significant changes in the serum levels of high density lipoprotein, low density lipoprotein and very low density lipoprotein of ARF rats when compared with negative control group. Rats which fed on diet fortified with GC seeds powder at 5% and 10% had lower mean values of LDL-C and VLDL-C compared with the positive control group confirmed by Behrouzian *et al.,* (2014) and Doke & Guha (2014). On the other hand, all treated groups (Curative & protective) with fortified diet at different levels of GC had higher mean values of HDL-C comparison with those of the positive control group.

Table 9: Effect of feeding acute renal failure rats on garden cress at different ratios on HDL-C; LDL-C and VLDL-C

Groups \ Parameters		HDL-C mg/dl	LDL-C mg/dl	VLDL-C mg/dl
Control (-ve)		$64.57^{a} \pm 2.103$	$58.14^{d} \pm 3.004$	$23.00^{d} \pm 0.362$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		$45.00^{d} \pm 5.325$	$158.14^{a} \pm 2.931$	$36.14^{a} \pm 1.361$
Curative groups	Control (+ ve) + 5% GC	$55.43^{c} \pm 6.209$	$78.43^{b} \pm 4.211$	$34.71^{b} \pm 0.421$
	Control (+ ve) + 10% GC	$63.57^{ab} \pm 4.113$	$70.86^{c} \pm 3.712$	$31.28^{c} \pm 0.835$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		$51.85^{d} \pm 3.324$	$125.00^{a} \pm 3.260$	$29.71^{a} \pm 1.210$
Protective groups	Control (+ ve) + 5% GC	$53.28^{c} \pm 3.572$	$101.03^{b} \pm 4.283$	$27.83^{b} \pm 0.262$
	Control (+ ve) + 10% GC	$61.33^{b} \pm 4.201$	$90.82^{c} \pm 2.384$	$24.85^{cd} \pm 1.014$

Effects of Garden Cress seed powder on kidney function of rats suffering from acute renal failure

Cisplatin induced renal toxicity with elevated biochemical markers such as serum urea and serum creatinine (Yadav *et al.,* 2010). Results presented in Table (10) revealed that there was significant difference ($P < 0.05$) between positive control group and other groups, (negative control, curative and protective groups) fed on GC seeds powder at 5% and 10%.

Serum uric acid:

Results indicated that oral administration of 5% and 10% *GC* powder reduced the uric acid level significantly. It was reduced in curative groups (*fed on 5 & 10% GC*) by 7.04% & 15.81%, and the concentration of uric acid was reduced in protective groups (*fed on 5 & 10% GC*) by 3.68% & 17.56% respectively. These findings are in agreement with Yadav *et al.,* (2009) revealed that nephrotoxicity induced by Cisplatin due oxidative stress and ethanolic extract of *Lepidium Sativum* seeds may be have nephroprotective and curative activity.

Serum urea nitrogen:

Urea nitrogen is a substance that is formed in the liver when the body breaks down protein. In healthy people, most urea nitrogen is filtered out by the kidneys and leaves the body in the urine. If the patient's kidneys are not functioning properly or if the body is using large amounts of protein, the serum urea nitrogen level will rise (Frey, 2007). On the other side, Yogesh et al (2010) observed that a single injection of Cisplatin caused reduction of glomerular filtration rate, which is accompanied by increase in urea nitrogen level indicating induction of acute renal failure.

Data presented in Table (10) showed that ARF rats had the higher values of urea nitrogen reached to 60.00±3.112 mg/ dl compared with negative control group 40.57±2.710 mg/ dl. In fact, there was significant difference ($P < 0.05$) between all groups (positive, negative and other all groups). Our results showed that the levels of urea nitrogen reached to 58.57±3.071 & 52.14±2.062 compared with positive control group 60.00±3.112 mg/ dl respectively in curative groups fed on *GC* seeds powder at 5% & 10%, while, with protective groups the levels of urea nitrogen reached to 60.16±0.480 & 56.57±0.351 compared with positive control group 64.28±0.172 mg/dl.

Table 10: Effect of feeding acute renal failure rats on Garden Cress at different ratios on kidney function

Groups \ Parameters		Uric acid mg/dl	Urea nitrogen mg/dl	Creatinine mg/dl
Control (-ve)		$2.94^{d} \pm 1.071$	$40.57^{d} \pm 2.710$	$0.492^{d} \pm 0.091$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		$5.82^{a} \pm 0.452$	$60.00^{a} \pm 3.112$	$1.525^{a} \pm 0.173$
Curative groups	Control (+ ve) + 5% GC	$5.41^{c} \pm 0.104$	$58.57^{b} \pm 3.071$	$1.004^{b} \pm 0.973$
	Control (+ ve) + 10% GC	$4.90^{b} \pm 0.274$	$52.14^{c} \pm 2.062$	$0.892^{c} \pm 0.471$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		$5.98^{a} \pm 0.663$	$64.28^{a} \pm 0.172$	$1.620^{a} \pm 0.691$
Protective groups	Control (+ ve) + 5% GC	$5.76^{b} \pm 0.810$	$60.16^{b} \pm 0.480$	$0.906^{b} \pm 0.665$
	Control (+ ve) 10% GC	$4.93^{c} \pm 0.552$	$56.57^{c} \pm 0.351$	$0.884^{c} \pm 0.453$

All results are expressed as mean ± SD.
Values in each column which have different letters are significantly different (p<0.05

Serum Creatinine:

Results in Table (10) showed that the creatinine increased with *Cisplatin injection* the values reached to 1.525±0.173mg/ dl compared with negative control group 0.492±0.091 mg/ dl. Also, data showed that the effects of feeding on basal diet containing GC seeds powder at 5% & 10% (curative groups), the levels of creatinine reached to 1.004±0.973 & 0.892±0.471 mg/ dl compared with positive control group 1.525±0.173mg/ dl respectively. While, with protective groups, the level reached to 0.906±0.665 & 0.884±0.453 mg/dl compared with positive control group 1.620±0.691 mg/dl.

From the previous studies by Tricia (2007) published that the creatinine is actually the broken down form of creatine that the kidneys process and dispose of normal measurement of creatine evaluates creatinine through blood tests. High creatinine levels may mean a person is severely dehydrated and also helps indicate if a person is experiencing kidney failure. In fact, our results confirmed that, GC seeds powder at 5% and 10% concentrations showed a protective effect against the acute renal failure induced by Cisplatin. Confirmed by Behrouzian *et al.,* (2014) and Doke & Guha (2014) they investigated that a dose of cisplatin induced increased creatinine level in serum and noticed that ethanolic extract of GC seeds may possess nephrocurative and nephrprotective activity. These finding can explain the possible effect of *GC* consumption at 5% and 10% on improving renal function for rats suffering from acute kidney disease.

Effects of Garden Cress seeds powder on liver enzymes of rats suffering from acute renal failure:

Results in Table (11) revealed that significantly higher ($P< 0.05$) value of AST & ALT with positive group, compared with those of negative control group, this may be referred into a direct excessive effect of Cisplatin on liver enzymes. Confirmed by Abd-El Fattah *et al.,* (2006) that the high levels of AST & ALT in serum are indicators for liver dysfunction. This significant increasing may be attributed mainly to the hepatotoxic effect of Cisplatin. In fact, administration of Curative & protective rats fed on basal diet containing GC seeds powder at 5% & 10% caused a significant reduction in the mean values of serum AST and ALT as compared to the positive control group, it appears from our results that high concentration of GC was safe and improves liver functions. Moreover, GC had shown significant hepatoprotective activity in several studies and can protect the liver from chemically induced damage confirmed by Vishwanath *et al.,* (2012) and Behrouzian *et al.,* (2014).

Table 11: Effect of feeding acute renal failure rats on garden cress at different ratios on liver function

Groups \ Parameters		AST (u/l)	ALT (u/l)
Control (-ve)		$20.85^{cd} \pm 1.634$	$14.00^{d} \pm 0.984$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		$25.14^{a} \pm 0.0976$	$19.85^{a} \pm 1.623$
Curative groups	Control (+ ve) + 5% GC	$22.85^{b} \pm 0.766$	$17.42^{b} \pm 1.129$
	Control (+ ve) + 10 GC	$21.28^{c} \pm 1.735$	$16.14^{c} \pm 0.745$
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		$24.00^{a} \pm 1.837$	$19.42^{a} \pm 1.612$
Protective groups	Control (+ ve) + 5% GC	$23.57^{b} \pm 1.082$	$18.50^{ab} \pm 0.574$
	Control (+ ve) + 10% GC	$22.33^{bc} \pm 1.608$	$17.28^{b} \pm 0.726$

Effects of Garden Cress seeds powder on Glutathione Reductase and Malondialdehyde of rats suffering from acute renal failure

MDA is a main product of lipid peroxidation, as a biomarker of oxygen free radicals; it has the potential not only to evaluate the extent of oxidative injury, but also to predict the potential efficiency of therapeutic strategies aimed at restricting the oxidative stress demonstrated by Yazdanparast *et al.,* (2007) and Halaby, *et al.,* (2013). As shown in Table (12) that the serum MDA level of the rats in the positive control group showed marked increase compared with rats in the negative control group (52.3±0.13 vs 33.4±0.21 (mg/dl). This may be referred into a direct excessive effect of Cisplatin on kidney function. However, rats in other groups experienced significantly ($p < 0.05$) less of rise in the serum MDA level as compared to positive control group.

The results given in Table (12) indicated that feeding ARF rats "curative groups" caused a significant reduction in the mean values of serum MDA; the values were 41.2±0.16 (for 5%) and 37.8±0.15 mg/dl (for 10%) as compared to the positive control group 52.3±0.13 mg/dl, respectively. Also, in the present study, with "protective groups" there were significant reduction in the mean values of serum MDA; the values were 46.8±0.07 (for 5%) and 42.7±0.84 mg/dl (for 10%) as compared to the positive control group 48.7±0.23 mg/dl, respectively.

Results of the present study (Table 12) showed that the level of glutathione (GSH) was significantly decreased ($p < 0.05$) in the serum of the rats in control positive group 6.21±0.08 mg/dl compared with those of negative control group (12.82±0.24 mg/dl) respectively. This may be referred into a direct excessive effect of Cisplatin on Glutathione Reductase confirmed by Yadav *et al.,* (2010) they published that after single dose injection of Cisplatin treatment decreased GSH level that indicates production of free radicals involvement of oxidative stress due to Cisplatin induced nephrotoxicity. Moreover, fed on basal diet containing GC seeds powder at 5% & 10% in "curative & protective groups" caused a significant increase in the mean values of serum GSH as compared to the positive control group. It may be suggested that the activities of the plant are due to its free radical scavenging activities and the rich content in antioxidants such as vitamin C, E and carotenoids, also phytochemical constituents such as (polyphenols and flavonoids) that have been reported to protect the body system against reactive oxygen species, as mentioned by Donno *et al.,* (2013).

According to Olorunnisola *et al.,* (2012) and Halaby, *et al.,* (2013) published that Glutathione helps in the regeneration of some important antioxidant vitamins such as C and E, depletion of GSH has been reported in apoptosis and many degenerative conditions. Confirmed by Behrouzian *et al.,* (2014) and Doke & Guha (2014) they noticed that the nephrocurative and nephroprotective activity of ethanolic extract of GC seeds was used against Cisplatin induced nephrotoxicity in adult male Wistar rats. Also, there was significant increase in glutathione level and decrease in lipid peroxidation in nephroprotective and curative test groups.

Table 12: Effect of feeding acute renal failure rats on garden cress at different ratios on Malondialdehyde and Glutathione Reductase of rats

Groups \ Parameters		(MDA) mg/dl	(GSH) mg/dl
Control (-ve)		33.4 ^{d} ±0.21	12.82 ^{a} ±0.24
Control (+ ve) Cisplatin injection (5mg/kg body wt.) from the first day		52.3 ^{a} ±0.13	6.21 ^{d} ±0.08
Curative groups	Control (+ ve) + 5% GS	41.2 ^{b} ±0.16	7.47 ^{c} ±0.05
	Control (+ ve) + 10% GS	37.8 ^{c} ±0.15	9.74 ^{b} ±0.03
Control (+ ve) Cisplatin injection (5mg/kg body wt.) after 28 days		48.7 ^{a} ±0.23	8.14^{d} ±0.05
Protective groups	Control (+ ve) + 5% GS	46.8 ^{b} ±0.07	8.96 ^{c} ±0.16
	Control (+ ve) + 10% GS	42.7 ^{c} ±0.84	9.05 ^{b} ±0.13

**Results are expressed as means ±SD.*
** Values in each column which have different letters are significantly different ($p<0.05$).*. Thermal Biolo., 35: 52–57.

Histopathological results

Kidneys:

A- “Curative groups:

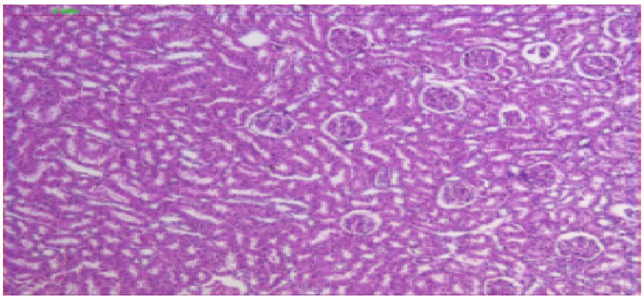


Photo 1: is a photomicrograph of a section of renal tissue of rat from negative control group fed on basal diet showed normal structure being formed of glomeruli embedded in between different types of tubules. These tubules are lined with cuboidal epithelium with rounded nuclei. (Hx. & E. X 100).

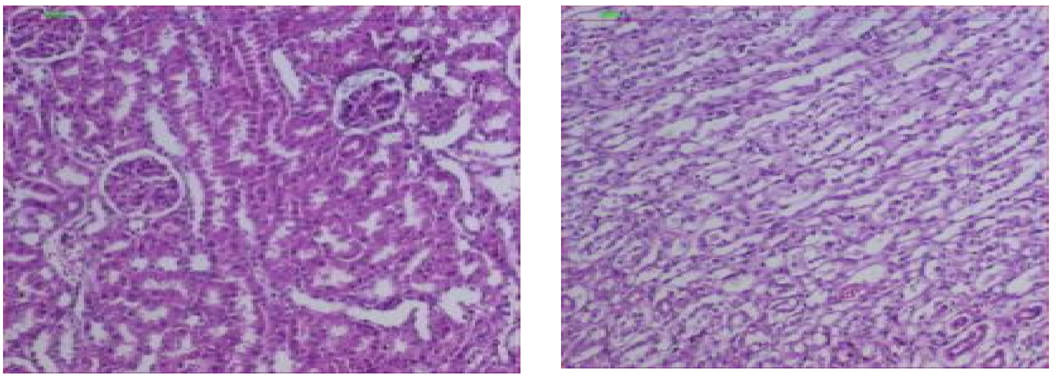


Photo 2: is a photomicrograph of a section of renal tissue of a rat from positive control group {Cisplatin injection (5mg/kg body wt.) from the first day) showed many tubules suffering from individual cell necrosis in their lining epithelium cellular infiltrate in the form of diffuse infiltration (left picture) associated with vacuolations in the distal tubules lining (right picture). (Hx. & E. X 200).

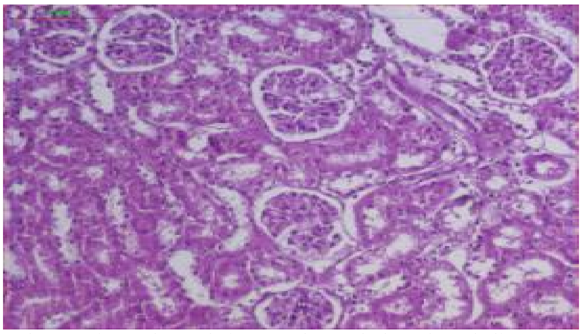


Photo 3: is a photomicrograph of a section of renal tissue of rat (fed on basal diet containing GC seeds powder at 5%) showed proximal tubules suffering from individual cell necrosis with some atrophic tubules and interstitial nephritis. (Hx. & E. X 200).

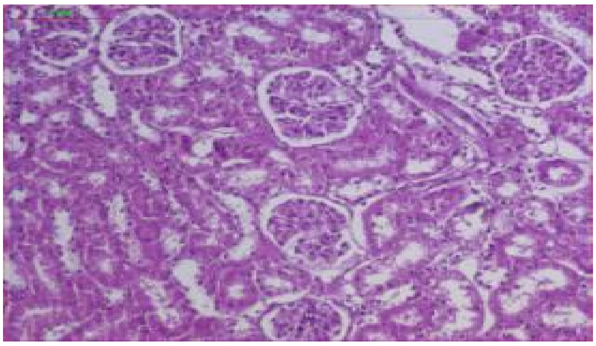


Photo 4: is a photomicrograph of a section of renal tissue of rat (fed on basal diet containing GC seeds powder at 10%) showed proximal tubules suffering from mild individual cell necrosis and minimal interstitial inflammation. (Hx. & E. X 200

B- "Protective groups":

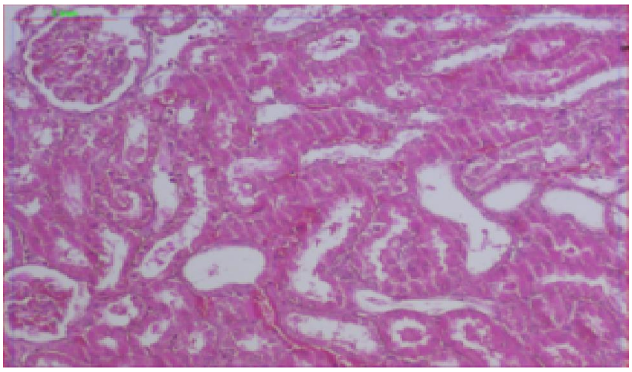
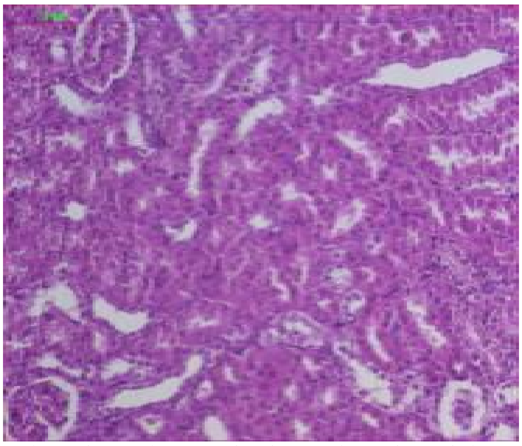


Photo 5: is a photomicrograph of a section of renal tissue of a control positive group (Cisplatin injection (5mg/kg body wt.) after 28 days) showed many tubules suffering from mild individual cell necrosis in their lining epithelium. Cellular infiltrate in the form of diffuse infiltration with vascular congestion. (Hx. & E. X 200)



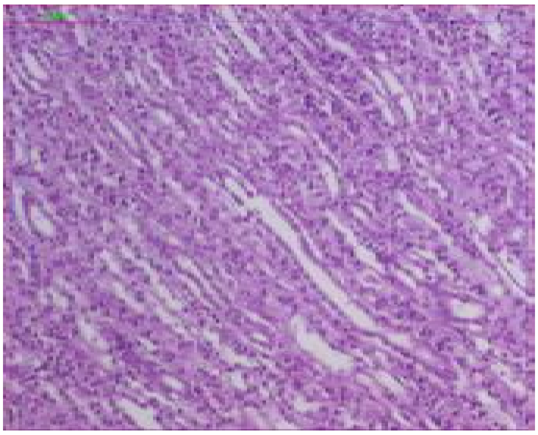


Photo 6: is a photomicrograph of a section of renal tissue of rat (fed on basal diet containing GC seeds powder at 5%, with Cisplatin injected after 28 days) showed many distal tubules suffering from severe vacuolar degeneration in their lining epithelium (right picture) and individual cell necrosis in the proximal tubular epithelium. Cellular infiltrate in the form of diffuse infiltration or focal aggregation with vascular congestion (left picture) (Hx. & E. X 200)

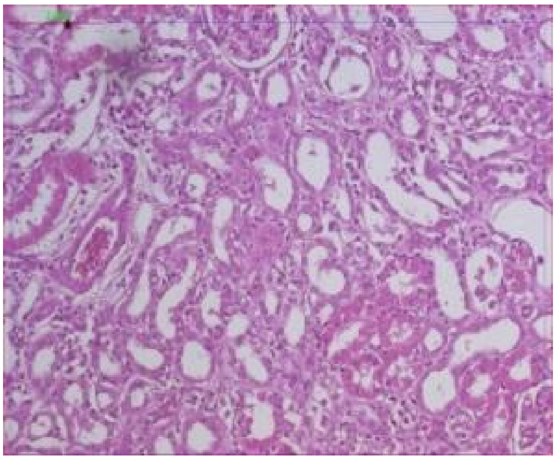


Photo 7: is a photomicrograph of a section of renal tissue of a rat (fed on basal diet containing GC seeds powder at 10%, with Cisplatin injected after 28 days) showed proximal tubules suffering from individual cell necrosiswith some atrophic tubules, vascular congestion and interstitial nephritis. (Hx. & E. X 200).

Conclusion

The current investigation concludes that *Garden Cress* seeds contained the various values of macro and micro-elements beside, flavonoid and polyphenolic compounds which might be responsible for its strong antioxidant capacity. Encouragement feeding ARF with *GC* seed to the our diets, which have the capability to improve the body weight gain, feed intake and feed efficiency ratio, with declining the serum uric acid, urea and creatinine, since it reduces the risk of acute kidney failure with the improving the activities of AST and ALT, as well as improvement of lipid profile.

Recommendation

Garden Cress seeds should be recommended for production on a commercial scale in the Egyptian meal, factories and medicines; such seeds have the capability to improve blood lipid levels as well as reducing hazards on kidney and liver function.

References

A.O.A.C., 2007. Official Methods of Analysis of the Association of Official Analytical Chemists. 18th Ed. Gaithersburg, Maryland, U.S.A.

Abd-El Fattah, S., Y. Abo Sree, M. El-Deeb, A. El-Bealawy and A. Helal, 2006. Toxicological studies on leucaena meal with two different dietary supplementation, on growth performance and appreciable residues of its toxic constituents (Mimosine and 3, 4- DHP) in milk of Egyptian goats. Journal of the Egyptian Vet. Med. Association, 66 (2): 83-99.

Agarwal, N., S. Sharma, 2013. Garden cress (*Lepidium sativum* L.). A non-conventional traditional plant item for food product .Indian journal of traditional knowledge, 12 (4): 699 - 706.

Al Hamedan, W., 2010. Protective Effect of *Lepidium sativum* L. Seeds Powder and Extract on Hypercholesterolemic Rats. Journal of American Science, 6 (11) 873- 879.

Allain,C., L. Poon, C. Chan, and W. Richmond, 1974. Enzymatic determination of total serum cholesterol. Clinical Chemistry, **20**: 470-475.

Al-Sheddi, E., N. Farshori, M. Al-Oqail, J. Musarrat, A. Al- Khedhairy and M. Siddiqui, 2013. Evaluation of cytotoxicity and oxidative stress induced by alcoholic extract and oil of *Lepidium sativum* seeds in human liver cell line HepG2 African Journal of Biotechnology. 12 (24), 3854-3863.

Armitage, P. and G. Berry, 1987. Statistical methods in medical research. Distributors, USA, Year Book Medical Publishers.

Bartels, H., M. Bohnzer and C. Heierli, 1972. This Week's Citation Classic . Clinical chemistry Acta, 37: 193.

Behrouzian, F., S. Razavi and G. Phillips, 2014. Cress seed (*Lepidium sativum*) mucilage, an overview. Bioactive Carbohydrates and Dietary Fibre, http://dx.doi.org/10.1016/j.bcdf.2014.01.001.

Bhasin, P., D. Bansal, O. Yadav and A. Punia, 2011. In Vitro Antioxidant Activity and Phytochemical Analysis of Seed extracts of *Lepidium sativum*- a medicinal herb. Journal of Bioscience And Technology. 2 (6): 410- 415.

Chapman, H. and P. Pratt, 1978. Methods of Analysis for soils. Plants and waters, Division of Agricultural sciences, university of California, Berkeley, USA, pp: 3043.

Chauhan, K., S. Sharma, N. Agarwal, S. Chauhan and B.vChauhan, 2012. A study on potential hypoglycemic and hypolipidemic effects of *Lepidium sativum* (Garden cress) in Alloxan induced diabetic rats. American Journal of PharmTech Research, 2: 522-535.

Diwakar, B., P. Dutta, B. Lokesh and K. Naidu, 2008. Bioavailability and metabolism of n-3 fatty acid rich garden cress (*Lepidium sativum*) seed oil in albino rats. Prostaglandins, Leukotrienes and Essential Fatty Acids, 78, 123-130.

Diwakar, B., P. Dutta, B. Lokesh, and K. Naidu, 2010. Physicochemical properties of Garden cress (*Lepidium sativum* L.) seed oil. Journal of the American oil chemists' society, 87, 539-548.

Doke, S. and M. Guha, 2014. Garden cress (*Lepidium sativum* L.) Seed - An Important Medicinal Source: A Review. Scholars Research Library. J. Nat. Prod. Plant Resour, 4 (1):69-80

Donno, D., G. Beccaro, M. Mellano, S. Canterino, A. Cerutti and G. Bounous, 2013. Improving the nutritional value of kiwifruit with the application of agroindustry waste extracts. Journal of Applied Botany and Food Quality 86: 11 – 15.

Drury, R.V. and E.A. Walligton, 1980. Carltons Histological techniques, 5thed, Oxford University Press, New York, Pronto, pp: 206 SY.

Fossati, P. and L. Prencipe, 1982. Serum Triglycerides Determined Colorimetrically with an Enzyme that Produces Hydrogen Peroxide. Clinical Chemistry, 28, 2077-2080.

Fossati, P., L. Prencipe, and G. Berti, 1980. Use of 3,5-dichloro-2-hydroxybenzene-sulfonic acid/4-aminophenazone chromogenic system in direct enzymatic assay of uric acid in serum and urine. Clinical chemistry 26: 227-231.

Frey, R.J., 2007. Blood urea nitrogen test health article. http://www.healthline.com/galecontent/blood-urea-nitrogen-test.

Friedewald, W., R. Levy and D. Fredrickson, 1972. Estimation of the concentration of low-density lipoprotein cholesterol without the use of the preparative ultracentrifuge. Clini. Chemi. 18: 499-502.

Gaafar, A., A. Morsi and H. Elghamry, 2013. Chemical, Nutritional and Biochemical Studies of Garden Cress Protein Isolate. Nature and Science 11 (2). 8-13.

Ghante, M., S. Badole and S. Bodhankar, 2011. Health benefits of garden cress (*Lepidium sativum* Linn.). In V. R. Preedy, R. R. Watson, & V. B. Patel (Eds.), Nuts and seeds in health and disease prevention (pp. 521-527). London: Elsevier Press.

Gokavi, S., N. Malleshi and M. Guo, 2004. Chemical composition of garde cress (*Lepidium sativum*) seeds and its fractions and use of bran as a functional ingredient. Plant Foods for Human Nutrition, 59, 105-111.

Goupy, P., M. Hugues, P. Biovin and M.J. Amiot, 1999. Antioxidant composition and activity of barley(Hordeum Vulgare) and malt extracts and of isolated phenolic compounds.J. Sci. Food Agric., 79: 1625-1634.

Halaby, M.S., M.H. Farag and A.Z. Mohammed, 2013. Influence of Kiwifruits on hypercholesterolemia in male albino rats. Egyptian J. of Nutrition and Health. Published by Society of Feeding mind, Combating malnutrition. 8 (1): 21 – 36.

Horwitz, W. and G. Latimer, 2007. Official methods of analysis. In: Association of Official Analytical Chemistry. 18th ed. Gaithersburg, AOAC International, USA. vol. xxix, 1v. (various pagings).
Indumathy, R., and A. Aruna, 2013. Free Radical Scavenging Activities, Total Phenolic and Flavonoid Content of *Lepidium sativum* (linn). International Journal of Pharmacy and Pharmaceutical Sciences. 5 (4).
ISO 5508, 1990.
ISO 5509, 2000.
Journal of chromatography, 1999. A 721: 247 – 259.
Journal of chromatography,2006. B 830: 41 – 46.
Kadam, P., K. Yadav, P. Shivatare, N. Narappanawar, A. Pande and M. Patil, 2012. *Lepidium sativum* Linn: An Ethnobotany and Phytopharmacological. International Journal of drug formulation and research. 3 (3), May-June.
Kasabe, P., P. Patil, D. Kamble and P. Dandge, 2012. Nutritional, Elemental Analysis and Antioxidant Activity of Garden Cress (*Lepidum sativum* L.) Seeds. Int J Pharm Pharm Sci, Vol 4, Issue 3, 392-395.
Lopes, M., S. Stone, S. Ellis and J. Collwell, 1977. Cholesterol determined in high denisty lipoprotein separated by three different methods. Clin. Chem.; 23 (5): 882.
Manohar, D., G. Viswanatha, S. Nagesh, V. Jain and H. Shivaprasad, 2012. Ethnopharmacology of *Lepidium Sativum* Linn (Brassicaceae): A Review. International Journal of Phytothearpy Reasearch. 2 (1).
Matthaus, B. and L. Angelini, 2005.Anti-nutritive constituents in oil seed crops from Italy, Ind crops Pro, 21: 89-99.
Mattila, K., S. Asikainen, J. Wolf, H. Jousimies-Somer, V. Valtonen and M. Nieminen, 2000. Age, dental infections, and coronary heart disease. J Dent Res 79:756-760.
Mora, L., L. deo antune, H. Francescato, and M. Bianchi, del, 2003. Pharmacol. Res. 47, 517- 522.
Moser, B., S. Shah, J. Winkler-Moser, S. Vaughn, and R. Evangelista, 2009. Composition and physical properties of cress (*Lepidium sativum* L.) and field pennycress (*Thlaspi arvense* L.) oils. Industrial Crops and Products, 30, 199–205.
Nayak, P.S., S.D. Upadhyaya and A.A. Upadhyaya, 2009. HTPLC densitometer determination of sinapic acid in chandrashur (*Lepidium sativum*). Journal of scientific research, 1 (1): 121-127.
Olorunnisola, O., G. Bradley and A. Afolayan, 2012. Protective Effect of *T. violacea* Rhizome Extract Against Hypercholesterolemia-Induced Oxidative Stress in Wistar Rats. Molecules 17, 6033-6045.
Oz, O., 2011. Effects of storage on composition of some vegetables of boiled and drying. Master Thesis. Ataturk University, Graduate School of Natural and Applied Science.
Patton, C.J. and , S.R. Crouch, 1977. Spectrophotometric and kinetics investigation of the Berthelot reaction for the determination of ammonia: Analytical Chemistry, v. 49, pp. 464-469.
Politeo, O., M. Jukic and M. Milos, 2006. Chemical composition and antioxidant activity of essential oils of twelve spice plants. Croat. Chem. Acta., 79 (4): 545-552.
Reeves, P., K. Rossow and J. Lindlauf, 1993. Development and testing of the AIN-93 purified diets for rodents: results on growth, kidney calcification and bone mineralization in rats and mice. J. Nutr. 123: 1923-1931.
Rehman, N., A. Khan, K. Alkharfy and A. Gilani, 2012. Pharmacological Basis for the Medicinal Use of *Lepidium sativum* in Airways Disorders. Research Article Evidence-Based Complementary and Alternative Medicine. Volume 2012, Article ID 596524, pp: 8.
Reitman, S. and S. Frankel, 1957. Colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. Amer. J. Clin. Pathol. 28: 56-63.
Ricardo, G., R. Cheyla, A. Borrego, F. Merino, M. Zamora, and E. Rojas, 2005. Lipid peroxides and antioxidant enzyme in cisplatin induced chronic nephrotoxicity in rats. Mediators Inflamm. 3: 139-143.
Sharma, S. and N. Agarwal, 2011. Nourishing and healing prowess of garden cress (*Lepidium sativum* Linn.)-A review. Indian Journal of Natural Products and Resources, 2 (3), 292- 297.
Subramonium, A. and P. Pushpangadan, 1999. Development of phytomedicines for liver diseases. Indian J Pharmacol. 1999; 31:166-75.
Tricia, E.C., 2007. What is Creatine. Copyright © by Conjecture Coporation. http://www.wisegeek.com/what-is-creatine.htm
Vishwanath J., P. G. Baddam, A. K. Boorgula and S. R. Jambula,(2012. A Review on Hepatoprotective Plants. International Journal of Drug Development & Research|July-September 2012 | Vol. 4: 5.
Yadav, Y., D. Srivastav, A. Seth, V. Gupta and S. Kuldeep, 2009. Nephroprotective and curative activity of *Lepidium Sativum* L. seeds in albino rats using cisplatin induced nephrotoxicity. Pharmacology on line 3: 640-646.
Yadav,Y., D. Srivastav, A. Seth, and V. Saini, 2010. Nephroprotective and curative activity of *Lepidium sativum* L. seeds in albino rats using cisplatin induced acute renal failure. Scholars Research Library. Der Pharma Chemica, 2 (4): 57-64.

Yazdanparast, R., A. Ardestani and S. Jamshidi, 2007. Experimental diabetes treated with Achillea santolina: effect on pancreatic oxidative parameters. J. Ethnopharmacol. 112:13–18.

Yogesh C.Y., D.N. Srivastav A.K. Seth and V. Saini, 2010. Nephroprotective and curative activity of *Lepidium sativum* L. seeds in albino rats using cisplatin induced acute renal failure. Scholars Research Library Der Pharma Chemica, 2 (4): 57-64.

Yogesh chand, Y., D. Srivastav, A. Seth, S. Vipin, R. Balaraman and K. Tejas, 2010. *Invivo* antioxidant potential of *Lepidium sativum* L. seeds in albino rats using cisplatin induced nephrotoxicity. Inter J Phytomed 2010; 2: 292-298.

Yogesh, C., J. Aijeet, D. Srivastava and J. Anurekha, 2011. Fracture healing activity of ethanolic extract of *Lepidium sativum* L. seeds in internally fixed rats' femoral osteotomy model International Journal of Pharmacy and Pharmaceutical Sciences, 3: 195-197.

Zaveri, M., N. Desai and V. Movaliya, 2011. Effect of Ocimum Basilicum on Cisplatin Models of Acute Renal Failure. ARPB, 1 (2).

Zia-Ul-Haq, M., S. Ahmad, L. Calani, T. Mazzeo, D. Del Rio, N. Pellegrini and V. De Feo, 2012. Compositional Study and Antioxidant Potential of *Ipomoea hederacea* Jacq and *Lepidium sativum* L. Seeds. Molecules 17, 10306-10321.