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Effect of Adding Nettle Leaves (*Uritca dioica* L.) powder on Basal Diet to Lower Diabetesin Rats

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STINGING nettle/common nettle (*Urtica dioica* L.) is known since ancient times as a wild source of food and an herbal medicine, but the plant remains underutilized. This investigation was carried to evaluate effect of adding nettle powder on basal diet to lowering diabetes in rats.

The results showed that nettle powder had contained a number of bioactive compounds as protein; crude fiber and ash were 30.54, 10.16and 15.24%, respectively. Nettle powder had contained relatively high amounts of bioactive compounds as tannin, total phenolic (TP) and total dietary fractions. Polyphenols fractions using HPLC had reported that the protocatechuic, quinic andcaffeic acids were the major compounds in phenolic acids, meanwhile, quercetin 3-O-glucoside, quercetin 3-O- rutinoside and kaempferol 3-O-glucoside were the major compounds in flavonoids compounds.

The results at the end of biological experimental for diabetic rats fed on basal diet supplemented with 5% and 10% nettle (*Urtica dioicaL.*) showed that nettle leaves powder may be help to maintain an ideal body weight and feed efficiency ratio. Diabetic rats were showed significant changes of relative weight of some internal organs. Diabetic ratsfed on 5 and 10% nettle powder were showed a significant enhancement of insulin secretion thereby decreasing the blood sugar level. The results from lipid profile showed significant reduction in the level of triglyceride, total cholesterol and LDL level in diabetic rats. The liver and kidney functions were improved in diabetic rats fed on 5 and 10% nettle (*Urtica dioica*) in basal diet.

From the above results it could be recommended that *Urtica dioica* leavespowder had contained highly amounts from protein, dietary fiber and natural antioxidant activity which might be useful in the development of new dietary supplement of versatile nature to improve lipid profile, liver and kidney functions in rats duo to its potent antioxidant property.

Keywords: *Uritca dioica*, Stinging nettle, Phytochemistry.

Introduction

Diabetes mellitus is considered as one of the five causes of death in the world (Joseph and Jini, 2013), approximately 150 million persons world wide are considered as diabetic patients according to World Health Organization , and this number may double by 2025 (Zhou et al., 2009). Alternative strategies to the current modern pharmacotherapy of diabetes mellitus are urgently needed because of the inability of existing modern therapies to control all the pathological aspects of the disorder, as well as the enormous cost and poor availability of the modern therapies for many rural populations in developing countries (WHO, 2002).

Anti-diabetic are scientifically motivated to discover newer, safer and affordable drugs that complement conventional strategies for management of diabetes. Therefore, scientific effectiveness for use of *Urtica dioica* has occurred for anti-diabetic activity. Moreover, *Urtica dioica* grows naturally in many parts of Africa with a wide variety use in traditional medicine and diet. (Mukundi et al., 2017).

Urtica dioica L. (common as stinging nettle) belongs to family *Urticaceae* and also, it is one of the most important genera and contains 30 species. In folk medicine, nettle has been used to treat iron deficiency anemia due to its high content of iron and also used to stop excessive menstrual

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bleeding, hematuria and nosebleeds. The root of nettle has been employed for treating asthma and the leaves of the nettle were used as laxative, diuretic and diabetics (Mikaeili et al., 2013).

Urtica dioica has a rich medicinal value and this plant is most commonly used to cure much of inflammatory disorders. In folk medicine this plantwas used for the treatment of arthritis and this plant is possessed antiasthmatic, antid and ruff, astringent, depurative, diuretic, galactogogue, haemostatic and hypoglycaemic activities in pre-clinical experiments. This plant has been long used in traditional medicine and food supplement by Native Americans and Europeans(Ahmed and Parsuraman, 2014).

Most animal studies have shown the beneficial effects of *Urtica dioica*indiabetes management. Human studies have demonstrated effectiveness of *Urtica dioica* as a potent stimulator of insulin release from β-cells and have shown protective effect on β-cells in diabetic rats. Moreover, it was inhibited intestinal absorption of glucose and inhibitory effects on the α-amylase activity are extra pancreatic mechanisms (Rashidi et al., 2013).

Stinging nettle leaves, not only add variety to the menu, but they are also valuable sources ofnutrients where they contribute protein, minerals, vitamins, fiber and phytochemicals (e.gphenolic compounds). Consumption of numerous types of wild edible plants especially greenleafy vegetables has been described as potentially beneficial for food and nutrition security aswell as to alleviate nutrition-related health problems (Naude, 2013 a,b).

The analyses of phenolic compounds of the *U. dioica* leaves were reported to contain caffeic acid and chlorogenic acid as reportd by Otles and Yalcin (2012), the total phenol content of nettle leaves ranged from 151- 1001 mg GAE/g and high content of chlorophyll and other pigments.

The aim of this investigation was carried out to evaluate the anti-diabetic effect of *Urtica dioica* L leaves powder. Chemical composition, total dietary fiber fraction and bioactive compound were determined. Fractionation of phenol and flavonoid compounds were assessed using HPLC. The effect of the leaves powder lowering lipid profile and improvement of liver and kidney functions in rats were evaluated.

Meaterials and Methods

Materials

Dried leaves of *Urtica dioica* (nettle) was purchased from Agriculture Seeds, Herbs and Medicinal Plants Company, Cairo – Egypt.

Kits of glucose, lipid parameters, liver function and kidney function were obtained from Bicon Diagnosemittel GmbH and Co. KG Hecke 8 made in Germany.

Casein, streptozotocin (STZ), saline, formalin and diethyl ether were obtained from Modern Lab Company in Cairo, Egypt.

Methods

Chemical proximate

Stinging nettle *Urtica dioica* leaves were immediately chemically analyzed for determination of moisture content, total protein, crude fat, crude fiber and ash content according to AOAC (2005). Total carbohydrate content was calculated by difference. Total dietary fibers, soluble and insoluble dietary fibers were determined in raw materials according to Prosky et al. (1992).

A number of functional properties of the above samples were also analyzed. The antioxidant activity as DPPH inhibition % was determined by the method described by Nuengchamnong et al. (2009). Total phenolic (mg GAE/g) was determined according to the method described by Ranganna (2001). Tannin as an anti-nutritional factor was determined according to AOAC (2005).

Extraction of Urtica dioica leaves

Ethanolic leaves extract was dried at 40◦C for three days and leaves powder was suspended in tubes with ethanol 96% incubated overnight at room temperature (25◦C), followed by centrifugation (13,000 revolutions per minute (rpm). The supernatant (the extract) was moved to another tube and kept at −20◦C until analysis for flavonoids and phenolic compounds using HPLC.

Quantitative determination of flavonoids by HPLC

Quantitative determination of flavonoids by HPLC analyses were performed according to Zuo et al. (2002). Each compound was identified by its retention time and by spiking with standards under the same conditions. The quantification of the sample was done by the measurement of the integrated peak area and the content was calculated using the calibration curve by plotting peak area against concentration of the respective standard sample.

Quantitative determination of phenolic compounds by HPLC

Phenolic compounds were determination by

HPLC according to the method of Goupy et al. (1999). Phenolic acid standard was dissolved in a mobile phase and injected into HPLC. Retention time and peak area were used to calculation of phenolic compounds concentration.

Biological experimental

Male albino rats (24 rats) weight ranging 150-200 g were purchased from the animal colony, Helwan farm, Vaccine and Immunity Organization, Ministry of Health, Cairo Governorate, Egypt. Animals were housed in individual cages with screen bottoms and fed ad *libitum* on a basal powdered diet appropriate for growing the rats which containing casein (20 % ≥ 85% protein), corn oil (8%), corn starch (31%), sucrose (31%), cellulose (4%), salt mixture (4%) and vitamin mixture (1%) according to Pell et al. (1992).

After feeding on basal diet for eight days, rats were divided into four groups. The first group (n= 6 rats) was fed on the basal diet rats and it was administered plain water for four weeks and considered as negative control. The second main group (18 rats) was fasted overnight and injected with streptozotocin (dissolved in 0.1M citric acid buffer and adjusted at pH 4.5) into the leg muscle (5mg /100g body weight) to induce diabetic and hyperglycemia rats according to Madar (1983). After 48 hr. of injection the second main group was divided into three sub groups (6 rats for each). The first one (6 rats) was continued to be fed on basal diet and considered as positive control. From the second and third subgroups (6 rats for each) were fed separately on basal diet fortified with 5 and 10% from *Urtica dioica leaves* powder. Each rat was weighted every two days and the food consumption was calculated.

Biological evaluation

Serum glucose was determined according to method describe byBrăslasu et al., (2007). Insulin wasestimated using Enzyme Linked Immunosorbent Assay (ELIZA) (King et al*.,* 2002).

The levels of serum total lipids, total cholesterol and triglycerides were determined according to knight et al. (1972), Allain et al. (1974) and Fossati and Prencipe (1982), respectively. High & low density lipoprotein-cholesterol in serum were determined according to Lopes-Virella et al. (1977) and Steinberg (1981), respectively. VLDL-ch was calculated as TG/5.

Serum liver enzymes activity, aspartate amino transferase (AST) and alanineamino transferase (ALT) were determined according to Bergmeyer , (1974). Alkaline phosphatase was determined according to Moss and Henderson (1999). Kidney functions as creatinine and urea were determined according toMurray and Kaplan (1984) andKaplan (1984).

Statistical analysis

The obtained data were exposed to analysis of variance. Duncan's multiple range tests at ($P \leq 0.05$) level was used to compare between means. The analysis was carried out using the PRO ANOVA procedure of Statistical Analysis System (SAS, 2004).

Results and Discussion

Chemical composition

The chemical analysis of *Urtica dioica* (nettle) leaves powder was carried out to determine the mean values of chemical composition on dry weight basis and the results are presented in Table 1. The moisture content of the leaves was 10.25%. These results occurred by Rutto et al. (2013) who found that the nettle plant contains relatively high level of moisture was 11%.

Stinging nettle powder showed higher amount of protein content 30.54%, while may be due to increase level of essential amino acids (Rutto et al., 2013). Crude fiber in the nettle powder is significantly higher than most of other plant foods.Nettle powder is one of the richest sources of crude fiber (10.16%). The level of crude fat is relatively low (3.68%).

Nettle powder contained 15.24% ash this may be caused *Urtica dioica* (nettle) is rich in minerals. Rutto et al. (2013) found that total ash content of nettle powder was 19.1%.Nettle powder contained the lowest amount of carbohydrate (30.13%), it shows the Nettle powder is much less glycemic.

Carbohydrate levels in the nettle powder decreases with increase in the protein content, fiber, ash, and fat as shown in Table 1. The results agree with the report given by Thapaliya (2010) and Palikhe (2012). Therefore, the current finding showed that the use of nettle powder in bakery products is likely to increase the protein, ash and fiber.

The result in the same table showed that the mean values of the soluble, insoluble and total dietary fiber contents in nettle powder were 4.26, 23.12 and 27.38%, respectively. Dietary fiber has been shown to have important health implications in the prevention for risk of chronic diseases such as cancer, cardiovascular diseases and diabetes mellitus (Mahattanatawee et al., 2006). Besides, dietary fiber has the ability to bind with bileacids and prevents its reabsorption in the liver,which inhibits cholesterol synthesis.Dietary fiber also enhances waterabsorption in the colon, thus prevent constipation (Trinidad et al., 2006).

Knipping et al. (2012) and Johnson et al. (2013) reported that nettle plant is rich in natural phenolic compounds and it play an important role in cancer prevention and treatment. Phenolic compounds from medicinal herbs and dietary plants had contained phenolic acids, flavonoids, tannins, coumarins, lignans, quinones, and others. Various bioactivities of phenolic compounds are responsible for their chemo preventive properties (e.g., antioxidant, anticarcinogenic, or antimutagenic and anti-inflammatory effects).

The result in the same table showed that the total phenolic of nettle powder had contented 172.21 mg GAE (Gaelic Acid Equivalent)/g. The increase in total phenol content could also probably be due to suppression of oxidation by antioxidants (Yamaguchi et al., 2001). The increase in antioxidant activity could be also linked to the antioxidants such as phenolic acids, flavonoids, tannins, curcuminoids, coumarins, lignans, etc. (Huang et al., 2010).

Tannin of nettle powder had contented 1.05 mg/100 g and also, the anti-oxidant activity was 67.5 % DPPH inhibition. Thapaliya (2010) reported that higher level of antioxidant activity (AA) was observed in nettle leaves powder.

From the above results it could be noticed that the nettle powder had contained relatively increased in bioactive compounds as tannin, total phenolic (TP), antioxidant activity (AA), this may be due to the nettle is rich source from bioactive compounds.

TABLE 1. Chemical composition and fiber fraction *Urtica dioica* (nettle) on fresh weight.

Chemical composition	Content fresh
Moisture (g/100g)	10.25±0.25
Total protein(g/100g)	30.54±0.13
Total fat (g/100g)	3.68±0.02
Crude fiber(g/100g)	10.16±0.27
Ash content(g/100g)	15.24±0.35
Total carbohydrates(g/100g)	30.13±0.61
Total dietary fiber(g/100g)	27.38±0.42
Soluble dietary fiber(g/100g)	4.26±0.04
Insoluble dietary fiber(g/100g)	23.12±0.38
Tannins (mg/100g)	1.05±0.01
Antioxidant activity (DPPH inhibition, %)	67.5±0.16
Total phenolic compounds (mg GAE/g)	172.21±0.34

The phenolic and flavonoids profiles in Urtica dioica extract (mg / g of dry extract)

Fractionation of phenolic acids and flavonoids compounds as mg/g were determined in *Urtica dioic*(nettle powder) andthe results are reported Table 2. From the results, it could be noticed that the major compounds in phenolic acids were protocatechuic, quinic andcaffeic acids (0.35, 0.31 and 0.25 mg/g, respectively). Vanillic, 5-O-caffeolylquinic, *p*-coumaric and ferulic acids were the manor phenolic acids compounds; meanwhile, gentisic acid was the lowest phenolic acids. The increase in total phenol content could be linked with extraction of theinsoluble phenolics such as condensed tannins and phenolic acidsbound to cellular polysaccharides or proteins (Giada, 2013).

The same table can report that the results from flavonoids compounds showed that the major compounds are quercetin 3-O-glucoside, quercetin 3-O- rutinoside and kaempferol 3-O-glucoside (0.65, 0.36 and 0.16 mg/g, respectively). As well as, the manor flavonoids compounds are kaempferol, rutin, hyperin (hyperoside) and quercitrin, whilst,isorhamnetin and isorhamnetin-3-O-rutinoside are the lowest flavonoid compounds. Nettle leaves were reported to contain phenolic acid (*e.g.* hydroxycinnamic acid, hydroxybenzoic acid), tannins and flavonoids (*e.g.* flavones, flavonols, iso-flavonols, anthocyanins, catechins, lignin) (Farag et al., 2013 and Orčić et al., 2014).

TABLE 2. The phenolic and flavonoids profiles in *Urtica dioica* extract (mg / g of dry extract).

mg / g extract	Flavonoids	mg / g extract	Penolic acids
0.012	Quercitrin	0.25	Caffeic acid
0.65	Quercetin 3-O-glucoside	0.023	*p*-Coumaric acid
0.36	Quercetin 3-O-rutinoside	0.011	Ferulic acid
0.071	Kaempferol	0.31	Quinic acid
0.16	Kaempferol 3-O-glucoside	0.072	Vanillic acid
0.0013	Isorhamnetin	0.35	Protocatechuic acid
0.005	Isorhamnetin-3-O-rutinoside	0.0063	Gentisic acid
0.063	Rutin	0.031	p-Hydroxybenzoic acid
0.051	Hyperin (hyperoside)	0.05	5-O-Caffeolylquinic acid

Effect of Urtica dioica (nettle) leaves on total food intake, body weight gain and feed efficiency ratio in diabetic rats

The data in Table 3 showed that the average increase in body weight gain was 42.6 g in negative control, while body weight gainwas decreased in diabetic rats group to 29.4 g as positive control fed on basal diet. Meanwhile, the rats fed on 5 and 10% *Urtica dioica* (nettle) leaves in basal diet, body weight gain was 34.4 and 32.2.This latter result may imply that the nettle leaves, by its hypoglycemic effect, may prevent the decrease of body weight. However, the exact mechanism of this process has not been identified completely (Ahmadi et al., 2015).

Whereas,the calculated data of feed efficiency ratio (FER) for rats fed on basal diet and different added from *Urtica dioica* (nettle) leaves was summarized in the same table. From the results, it can be reported that the control positive and negative for food efficiency ratio (FER) were 0.08 for both , whilst,t he rats fed on 5 and 10% *Urtica dioica* (nettle) leaves in basal diet,had the same results. From The results showed that there is no significant change in FER.

TABLE 3. Effect of nettle leaves on total food intake, body weight gain and feed efficiency ratio in diabetic rats (mean ± SD, n=6).

Groups	FI (g)	BWG (g)	FER
Control negative -ve	504± 39.11[a]	7.12[a]42.6±	0.01[a]0.08±
Control positive+ve	38.67[c]354±	5.38[c]29.4±	01c[a]0. 0.08±
Diabetic + nettle leaf (5%)	34.20[b]408±	4.27[b]34.4±	01[a]0. 0.08±
Diabetic + nettle leaf (10%)	53.66[c]366±	32.2±460[b]	0.01[a]0.08±

Means in the same column with completely different letters are significantly different at $p<0.05$.

Effects of Urtica dioica (nettle) leaves on the relative weight of some internal organs in diabetic rats

The results from Table 4 showed that the liver weight was 2.43g in negative control and decreased in the group positive control to 2.10 g and it was slightly increased (2.40 and 2.42g) in the groups that fed on 5 and 10% *Urtica dioica* (nettle) leaves in basal diet, respectively than control negative. The results showed that the weight kidney was 1.09g in negative control, and decreased in positive control 1.01g. Whereas the rats groups fed on 5 and 10% *Urtica dioica* (nettle) leaves in basal diet were 1.06 g and 1.08 g, respectively. Moreover, the pancreas weight in positive control was the lowest 0.20 g. These results revealed that hyperglycemia and diabetes in rats caused significant changes of relative weight of some internal organs.

TABLE 4. Effects of nettle leaves on the relative weight of some internal organs in diabetic rats (mean± SD, n=6).

Groups	Liver (%)	Kidneys (%)	Pancreas (%)
Control negative -ve	2.43±0.1[a]	0.21[a]1.09±	0.05[a]0.29±
Control positive+ve	2.10±0.2[b]	0.09[b]1.01±	0.07[b]0.20±
Diabetic + nettle leaf (5%)	2.40±0.2[a]	0.19[a]1.06±	0.03[a]0.27±
Diabetic + nettle leaf (10%)	2.42±0.4[a]	0.05[a]1.08±	0.01[a]0.29±

Means in the same column with completely different letters are significantly different at $p<0.05$.

Effects of Urtica dioica(nettle) leaves on insulin and glucose in serum of diabetic rats

Insulin and glucose were determined in serum diabetic rats and the results are reported in Table 5. From the results it could be observed that the control positive was the highest in serum glucose (248.14 mg/dl) and the lowest in insulin was 0.92 µU/mL meanwhile, the results from control negative was oppositely. Moreover, the diabetic rat fed on 5 and 10% *Urtica dioica* leaves in basal diet were increased in insulin (1.94 and 2.22 µU/mL) and decreased in glucose was 140 and 120 mg/dl compared with control positive. These results agree with Bnouham et al. (2003) who found that the *Urtica dioica* leaves has shown asignificant glucose lowering effect against diabetes in rats and also, it was administered in perfused islets of langerhans both in normal and streptozotocin induced diabetes in rats which showed a significant enhancement of insulin secretion thereby decreasing the blood sugar level (Farzami et al., 2003).

TABLE 5. Effects of nettle leaves on insulin and glucose in serum of diabetic rats(mean± SD, n=6).

Groups	Insulin (µU/mL)	Glucose (mg/dl)
Control negative -ve	2.28± 0.49[a]	117± 11.69[c]
Control positive+ve	0.46[c]0.92±	248±14.35[a]
Diabetic + nettle leaf (5%)	0.15[b]1.94±	140±8.34[b]
Diabetic + nettle leaf (10%)	0.29[a]2.22±	120± 9.44[c]

Means in the same column with completely different letters are significantly different at $p<0.05$.

Effect of Urtica dioica(nettle) leaves on lipid profile in serum of diabetic rats

Triglyceride and total cholesterol were determined in diabetic' rats fed on *Urtica dioica* leaves and the results are tabulated in Table 6. The result illustrated that the triglyceride, total cholesterol, LDL and VLDL were the highest in control positive (222, 220, 89 and 55 mg/dl, respectively) and the lower in control negative was 110, 96, 33 and 19 mg/dl, respectively. Moreover, the rats fed on *Urtica dioica* leave at 5 and 10% in basal diet were decreased in control positive and slightly increased in control negative. Meanwhile, LDL was the highest in control diabetic rats (89 mg/dl) and the lowest in the control negative was 33mg/dl) LDL levels in diabetics rats fed on *Urtica dioica* leaves at 5 and 10% in basal diet were decreased compared with control positive and slightly increased compared with control negative. These results are similar to several studies have demonstrated that the uniquely high level of polyphenols in *Urtica dioica* leaves may play an important role in contributing to the health benefit such as lowering cholesterol and diabetes level.

Daher et al. (2006) observed that the *Urtica dioica* leaves powder has lowered the levels of lipids and lipoproteins in blood. The significant decrease was found in lipid profile as total cholesterol, cholesterol fractions and LDL/HDL ratios via lower concentrations of LDL and plasma total apo-protein B. Moreover, the ethanolic extract of the plant at dose 100 and 300 mg/kg has shown significant reduction in the level of total cholesterol and LDL level in hypercholesterolemia and diabetics rats (Avci et al., 2006 and Nassiri-Asl et al., 2009).

TABLE 6. Effect of nettle leaves on lipid profile in serum of diabetic rats (mean± SD, n=6).

Groups	TC (mg/dl)	TG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
Control negative -ve	110± 19[b]	96± 23[b]	57± 6.1[a]	33± 10[ab]	19± 5.6[c]
Control positive+ve	222± 22[a]	220± 25[a]	23± 2.5[b]	89± 14[a]	44± 4.2[a]
Diabetic + nettle leaf (5%)	115± 8[b]	120± 11[b]	54± 4.1[a]	51± 11[b]	24± 3.1[b]
Diabetic + nettle leaf (10%)	116± 10[b]	100± 13[ab]	50± 3.7[a]	42± 12[ab]	20± 2.8[c]

Means in the same column with completely different letters are significantly different at $p<0.05$.

Effects of Urtica dioica (nettle) leaves on liver functions in serum of diabetic rats

The serum alanine (ALT) and aspartate (AST) transaminoferase values of rats fed on different diets under investigation during the experimental period are summarized in Table 7. The results showed that the aspartate transaminoferase (AST) activity in positive control group was 43 U/L and also it was significant increase in serum alanine (ALT) transaminoferase activity which elevated level 95U/L compared with negative control. Moreover, the diabetics rat fed on 5 and 10% *Urtica dioica* leaves in basal diet were decreased in AST (27 and 26U/L) and decreased in ALT was 45 and 40 U/L compared with control positive.

The results in the same table observed that the alkaline phosphatase (ALP) was parallel to the results from aspartate and alanine transaminoferase activity. These results are consistent with Kandis et al. (2010) who showed that the *Urtica dioica* leaves presentation hepatic damage created with ischemia reperfusion and it exhibited liver protection effect by increasing the activity of paraoxonase, arylesterase and liver tissue catalase activity**.**

Hepatoprotection is the ability to prevent damage to the liver, prevent the liver affections prophilactically andmaintains balance in liver enzymes. The *Urtica dioica*leaves extract of plant has shown maximum hepatoprotective activity at dose 400 mg/kg to lowering level of serum alanine transaminase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) (Ahangarpour et al., 2012 and Kataki et al., 2012).

TABLE 7. Effects of nettle leaf on liver functions in serum of diabetic rats (mean± SD, n=6).

Groups	ALT (U/L)	AST (U/L)	ALP (g/dl)
Control negative -ve	39± 5.02[b]	25± 5.02[b]	2.86± 0.38[c]
Control positive+ve	95± 25.2[a]	43± 8.11[a]	4.58± 0.29[a]
Diabetic + nettle leaf (5%)	45± 9.23[b]	27± 7.09[b]	3.26± 0.16[b]
Diabetic + nettle leaf (10%)	40± 7.85[b]	26± 8.41[b]	3.00± 0.86[b]

Means in the same column with completely different letters are significantly different at $p<0.05$**.**

Effects of Urtica dioica(nettle) leave on kidney functions in serum of diabetic rats

Results present in Table 8 showed that the values of serum urea of rats under investigation during the experimental periods. The results indicated that, the serum urea in negative control group was decreased to 26.4 mg/dl than positive control group showed a significant increased to 42.8 mg/dl. It could be observed that groups that have fed on *Urtica dioica* leaves at 5 and 10% in basal diet were reduced values of serum urea 35.6 and 28.6 mg/dl compared with positive and negative control.

The results in the same table showed that the relative decreased in serum creatinine was observed in the group of rats fed on basal diet was 0.64 mg/dl in negative control and increased to 0.92mg/dl in diabetic positive control. Meanwhile the rat groups fed on diet had contained *Urtica dioica* leaves at 5 and 10% in basal diet were decreased to 0.77 and 0.70 mg/dl compared with positive control. Treasure (2003) showed that nettle leaves can be quite effective for reducing serum creatinine levels in patients with chronic renal failure. Moreover, nettle leaves appear to also be hepatoprotective based on studies in rodents and also, the clinical experimental are could be definitely warranted with this completely safe herbal medicine (Kanter et al., 2003).

TABLE 8. Effects of nettle leaf on kidney functions in serum of diabetic rats (mean ± SD, n=6).

Groups	Urea (mg/dl)	Creatinine (mg/dl)
Control negative -ve	26.4± 2.70[ab]	0.64± 0.11a[b]
Control positive+ve	42.8± 11.65[a]	0.92± 0.10[a]
Diabetic + nettle leaf (5%)	35.6± 3.28[b]	0.77± 0.10[b]
Diabetic + nettle leaf (10%)	28.6± 5.94[ab]	0.70± 0.15[ab]

Means in the same column with completely different letters are significantly different at $p<0.05$.

Conclusions

Urtica dioica (nettle) powder has contained the highest amount of protein, crude fiber, ash content and relatively low in carbohydrate were 30.54, 10.16, 15.24 and 40.38%, respectively. Besides, it has excellent health enhancing functional properties; and also, it has much higher level of tannin content, total phenolic acid and antioxidant activity, these components had functional properties may play important role in lowering diabetes, hypochlosterolemic and potentially cancer prevention. Stinging nettle (*Urtica dioica*) is a common herb has a great medicinalvalue such as relieve of lowering glucose in blood, lowering lipid profile and improvement of liver and kidney functions.

References

Ahangarpour A., Mohammadian M. and Dianat M. (2012) Antidiabeticeffect of hydroalcohalic *urtica dioica* leaf extract in male rats withfructose-induced insulin resistance. Iran J Med Sci 2012; 37:181-6.

Ahmed MKK and Parsuraman, S. (2014) *Urtica dioica* L.,(*Urticaceae*): A Stinging Nettle, *Systematic Reviews in Pharmacy,* Vol 5, Issue 1, Jan-Dec, 2014, 6-8

Ahmadi, S., Awliaei, H., Haidarizadeh, M. and Rostamzadeh, J. (2015) The effect of ethanolic extract of *urtica dioica* leaves on high levels of blood glucose and gene expression of glucose transporter 2 (glut2) in liver of alloxan-induced diabetic mice.*Gene Cell Tissue.* 2015 July; 2 (3): e30355

Allain, C.C.; Poon, C.S.G; Chan, Richmond ,W. and Fu, P.C. (1974) Enzymatic determination of serum cholesterol. Clin. Chem., 20: 470-475.

AOAC. (2005) *Official Methods of Analysis* of AOAC International, 18th ed. AOAC International, Washington,DC.

Avci G., Kupeli E., Eryavuz A., Yesilada E., Kucukkurt, I. (2006) Antihypercholesterolaemic and antioxidant activity assessmentof some plants used as a remedy in Turkish folk medicine.*J Ethnopharmacol.* 2006;107:418-23.

Bergmeyer, U.H. (1974) *Methods of Enzymatic Analysis,* 3rd ed, Verlag Chemie Press Germany. Vol. II and IV. pp. 864, 1786.

Bnouham M., Merhfour F.Z., Ziyyat A., Mekhfi H., Aziz M. and Legssyer A. (2003) Antihyperglycemic activity of the aqueous extract of *Urtica dioica*. *Fitoterapia,* 2003; 74:677-81.

Brăslasu, M.C ; Brăslasu, E.D. and Brădăłan, C. (2007) Experimental studies regarding the diabetes mellitus induced in white wistar rats. *Lucrări Stiinłifice Medicină Veterinară,* **11**,109–116.

Daher C.F., Baroody K.G. and Baroody G.M. (2006) Effect of *Urtica dioica*extract intake upon blood lipid profile in the rats. *Fitoterapia,* 2006;**77**,183-8.

Farag, M.A., Weigend, M., Luebert, F., Brokamp, G. and Wessjohann, L.A., (2013) Phytochemical,phylogenetic, and anti-inflammatory evaluation of 43 Urtica accessions (stinging nettle) based on UPLC-Q-TOF-MS metabolomic profiles. *Phytochemistry,* **96**, 170–183.

Farzami B., Ahmadvand D., Vardasbi S., Majin F.J., Khaghani Sh. (2003) Induction of insulin secretion by a component of *Urtica dioica*leave extract in perfused Islets of Langerhans and its *in vivo* effectsin normal and streptozotocin diabetic rats. *J. Ethnopharmacol.* 2003;89:47-53.

Fossati, P. and Prencipe, L. (1982)The determination of triglyceride using enzymatic methods. *Clin. Chem.,* **28** , 2077.

Goupy, P., Hugues, M.m Biovin, P. and Amiot, M. J. "Antioxidant composition and activity of barley *(Hordeum Vuigare*) and malt extracts and of isolated phenolic compounds", *J. Sci. Food Agric,* vol. **79**, pp. 1625 -1634, 1999.

Huang, W. Y., Cai, Y. Z. and Zhang,Y. (2010) Naturalphenolic compounds from medicinal herbs and dietaryplants: potential use for cancer prevention. *Nutrition and Cancer* **62**,1–20.

Johnson, T. A., Sohn, J., Inman,W. D., Bjeldanes, L. F. and Rayburn, K. (2013) Lipophilic stinging nettle extracts possess potent anti-inflammatory activity, are not cytotoxic and may be superior to traditional tinctures fortreating inflammatory disorders. *Phytomedicine,* **15,** 20,143–147.

Joseph, B. and Jini, D. (2013) Anti-diabetic effects of *Momordica charantia* (bittermelon) and its medicinal potency. *Asian Pac, J, Trop, Dis,* **32,** 93-102.

Kandis H., Karapolat S., Yildirim U., Saritas A., Gezer S., Memisogullari R. (2010) Effects of *Urtica dioica* on hepatic ischemia-reperfusion injury inrats. *Clin.* (Sao Paulo) 2010; **65,**1357-61.

Kanter M., Meral I. and Dede S. (2003) Effects of *Nigella sativa* L. and *Urticadioica* L. on lipid peroxidation, antioxidant enzyme systems and some liverenzymes in CCl4-treated rats. *J. Vet. Med. A Physiol. Pathol. Clin. Med.* 2003;**50,**264–268.

Kaplan, A. (1984) *Urea. Clin Chem.,* 1257- 1260 and 437 and 418.

King et al., M.J. King, I. Badea, J. Solomon, P. Kumar, K.J. Gaspar, M. Foldvari (2002) Transdermal delivery of insulin from a novel biphasic lipid system in diabetic rats. *Diabetes Technology &Therapeutics,* **4** (4) (2002), pp. 479-488

Kataki, M.S., Murugamani, V., Rajkumari , A., Mehra, P.S., Awasthi, D., Yadav, R.S. (2012) Antioxidant, hepatoprotective and anthelmintic activities of methanol extract of *Urtica dioica* L. Leaves. *Pharm. Crops,* 2012;**3,**38-46.

Knight, J.A., Anderson, S. and Rowle, J.M. (1972) Chemical bases of the sulpfo-phospho vanillin reaction for estimating total serum lipids. *J. Clin. Chem.,* **18,** 199.

Knipping, K., Garssen, J. and Van't Land, B. (2012) An evaluation of the inhibitory effects against rotavirus infection of edible plant extracts. *Virol. J.* **9,**135–137.

Lopes-Virella, M.F. , Stone, S. , Ellis, S. and Collwel, J.A. (1977) Cholesterol determination in high density lipoproteins separated by three different methods. *Clin. Chem.,* **23** (5),882-884.

Mader, Z. (1983) Effect of brown rice and soybean dietary fiber on the control, glucose and lipid metabolism in diabetic rats. *Am. M. J. Clin. Nutr.,* 388-393.

Mahattanatawee, K., Manthey,J.A., Luzio, G., Talcott,S.T., Goodner, K. and Baldwin,E.A. (2006) Total antioxidant activity and fiber content of selectFlorida-grown tropical fruits. *J. Agric. Food Chem.,* **54,** 7355-7363.

Mikaeili A., Karim I., Modares M., Bagherinasab Z. (2013) Assessment of Antidermatophytic Activities of *Urtica dioica* L. against *Microsporum canis* in a Guinea Pig model. *Trop. J. Pharm. Res.* **12,** 997-1002

Moss, D.W. and Henderson, A.R. (1999**)** Clinical enzymology In: Burtis CA, Ashwood ER, Editors, *Tiets Textbook of Clinical Chemistry,* W.B. Saunders Company, Philadelphia 1999; 617-721.

Murray, R. L. and Kaplan, A. (1984) *Creatinine. Clin Chem.,* 1261- 1266 and 418.

Mukundi, M.J., Mwaniki, N.E.N., Piero, N.M., Murugi, N.J. and Kelvin, JK. (2017) Potential Anti-diabetic Effects and Safety of Aqueous Extracts of Urtica dioica - Collected from Narok County, Kenya. *Pharm Anal Acta,* **7**: 548. doi: 10.4172/2153- 2435.1000548

Nassiri-Asl, M., Zamansoltani, F., Abbasi, E., Daneshi, M.M., and Zangiv, A.A. (2009) Effects of *Urtica dioica* extract on lipid profilein hypercholesterolemic rats. *Zhong Xi Yi Jie He Xue Bao.* 2009;**7**,428-33.

Naude, C.E. (2013a) Would an increase in vegetable and fruit intake help to reduce the burden of nutrition-related disease in South Africa? An umbrella review of the evidence. *South African Journal of Clinical Nutrition,* **26,** 104–114.

Naude, C.E. (2013b) Food-based dietary guidelines for South Africa: The 'Eat plenty of vegetables and fruit every day'. South African

Journal of Clinical Nutrition, **26,** 46–56.

Nuengchamnong, N., K. Krittasilp, and Ingkaninan, K. (2009**)** Rapid screening and identification of antioxidants inaqueous extracts of Houttuyniacordata using LC–ESI–MScoupled with DPPH assay. *Food Chemistry.* **117,**750–756.

Orčić, D., Francišković, M., Bekvalac, K., Svirčev, E., Beara, I., Lesjak, M., Mimica-Dukić, N. (2014) Quantitative determination of plant phenolics in *Urtica dioica* extracts by high performance liquid chromatography coupled with tandem mass spectrometric detection. *Food Chemistry,* **143,** 48–53.

Otles, S., Yalcin, B. (2012) Phenolic compounds analysis of root, stalk, and leaves of nettle. *The Scientific World Journal,* 2012, 1–12.

Palikhe, M. (2012) Preparation of biscuit incorporating nettlepowder and its quality evaluation. *B. Tech. (Food) Thesis.* College of Applied Food and Dairy Technology, Purwanchal University, Nepal.

Pell, J.D., Gee, J.M., Wortley, G.M. and Johnson, I.T. (1992) Both dietary corn oil and guar gum stimulate intestinal crypt cell proliferation in rats, by independent but potentially synergistic mechanisms. *J. Nutr.,* **122,** 2447–2456.

Prosky, L., Asp, N.G. Schweizer, T.F., Vries, J.W. and Furda, I. (1992) Determination of insoluble andsoluble dietary fiber in foods and food products:Collaborative study. *J. AOAC Int.,* **75,** 330-367.

Ranganna, S. (2001) *Handbook of Analysis and Quality Control for Fruit and Vegetable Products.* Tata McGraw-Hill publishing Co. Ltd, New Delhi.

Rashidi, A.A., Mirhashemi, M.S., Taghizade, M., and Sarkhail, P. (2013) Iranian medicinalplants for Diabetes mellitus. A systematic review. *Pak. J. Biol. Sci.* **16,** 401-411.

Rutto, L.K., Xu,Y., Ramirez, E. and Brandt. M. (2013) Mineral properties and dietary values raw and processedStinging nettle (*Urtica dioica* L.). *Int. J. Food Sci.* 2013.doi: 10.1155/2013/857120.

SAS Institution (2004) SAST user statistics, var 6.04, 4th ed. SAS. Institution.Cary, N. C.

Steinberg, D. (1981) Metabolism of lipoproteins at the cellular level in relation to atherogenesis In lipoproteins.Atherosclerosis and Coronary *Heart Disease,* **1**(2), 31-48.

Thapaliya, P. (2010) Effect of different blanching conditions,drying methods and temperatures and retention ofvitamin C on *sisnu* powder. *M. Tech (Food) Thesis,*Tribhuvan University, Nepal.

Treasure J. (2003) Urtica semen reduces serum creatinine levels. *J. Am. Herbalists Guild* 2003;**4,**22–25.

Trinidad, T.P., Mallillin, A.C., Valdez, D.H., Loyola, A.S., Askali-Mercado, F.C., Castillo,J.C. Encabo, R.R., Masa, D.B., Maglaya, A.S. and Chua, M.T. (2006) Dietaryfiber from coconut flour: A functional food. *Innovative Food Sci. and Emerging Technol.,* **7,** 309-317.

WHO (2002) The World Health Organization (WHO) (2002) WHO launches the first globalstrategy on traditional medicine: Press release WHO/38

Yamaguchi, T., Mizobuchi, T., Kajikawa, R. (2001) Radical-scavenging activity of vegetables and the effect of cooking on their activity. *Food Science and Technology Research,* **7,** 250–257.

Zhou T., Luo D., Li X. and Luo Y. (2009) Hypoglycemic and hypolipidemic effects offlavonoids from lotus (*Nelumbo nuficera* Gaertn) leaf in diabetic mice. *J. Med Plant Res.* **3,** 290-293.

Zuo, Y., Chen, H. and Deng, Y. (2002) “Simultaneous Determination of Catechins Caffeine and Gallic acids in Green, Oolong, Black and Purer Teas using HPLC with a Photodiode Array Detector”, *Talanta,* vol. **57,** pp. 307-316, 2002.

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