Evaluation of Anti-Obesity Effect of Different Levels of Purslane in Albino Rats

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Abstract:

Obesity, one of the worldwide health problems, it considered increase in fat mass accumulation and body weight gain. The main aim of this study was to find the anti-obesity effect of Purslane *(Portulaca oleracea)* seeds .Thirty (30) adult male albino rats (Sprague-Dawley strain), were distributed into five main groups (6 rats each) as follows: Group (1) negative or normal group (-Ve): Fed on basal diet only as a control negative. Group (2) positive group (+Ve): were fed on high fat diet (HFD) as a control positive. Group (3): were fed on HFD supplemented with the grinded Purslane seeds 5%. Group (4): were fed on HFD supplemented with the grinded Purslane seeds 10%. Group (5): were fed on HFD supplemented with the grinded Purslane seeds 15%. Results indicated that supplementation with Purslane caused a significant decrease ($P<0.05$) in body weight of obese rats compared to the control positive group. Moreover, serum glucose, leptin, cholesterol, triglycerides, LDL-c, VLDL-c, urea, uric acid, creatinine, AST, ALT and ALP were significantly decreased, while serum HDL-c was significantly increased.

Key words: Purslane seeds, body weight, blood lipids, kidney functions, and liver enzymes

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Introduction

Obesity, one of the worldwide health problems, it is considered increase in fat mass accumulation and body weight gain. This illness results from the rise of high caloric food intake and reducing physical activity. Obesity is the chief risk factor for metabolic syndrome, which is connected with increased risk of several types of chronic diseases, for example hypertension, hyperlipidemia, cardiovascular diseases, fatty liver and insulin resistance. Overweight people are responsible for the deaths of at least 2.8 million individuals every year, according to the World Health Organization (WHO) **(AL-Faris, 2014)**. Egypt has a very high obesity rate. Between 61% and 70% of adults aged 20 and up are considered overweight or obese (BMI≥25 kg/m²). This works out to 76% for girls and 65% for boys in the age bracket of 15 and up. Concerning obesity specifically, between 18% and 22% of men and 39% to 48% of women are overweight **(WHO, 2010; IASO, 2012). Ali *et al.*, (2014)** took 500 milligrams of P. oleracea seed powder twice day for a month. Cholesterol, LDL-c, HDL-c, and TG levels, among other biochemical indicators, were assessed both before and after the study's initiation and termination. **Ramadan *et al.*, (2017)** investigated whether extract from Portulaca oleracea could protect diabetic rats' pancreas from damage and induce hypoglycemia. Rats given Regla extract lost significantly less weight than control rats, according to the findings. Thus, the aim of this study was to find the anti-obesity effect of Purslane seeds .

Martial and Methods

Materials:

Purslane *(Portulaca oleracea)* plant seeds were obtained from the Agriculture Research Center in Cairo. The kits were obtained from bio diagnostics firm in Cairo, Egypt. El-Gomhoriya Pharmaceutical of Cairo, Egypt, was supplied the chemicals, vitamins, and minerals, including casein, cellulose, choline chloride, D-L methionine, and others. In this study, thirty albino male Sprague-Dawley rats, each weighing 220±10 g, were procured from the Farm of Experimental Animals in Helwan, Egypt.

Ethical Approval

All experiments of the study were ethically approved by the Scientific Research Ethics Committee From the University of Alexandria, Animal Ethics Committee, Faculty of Medicine (Approval no. 05- 2025-01, SREC0307051).

Experimental design:

Thirty male albino Sprague-Dawley rats, with an average weight of 220±10 g, were kept in group cages with controlled temperatures (22-24°C) and lighting (12-hour light cycle beginning at 6 AM) for a minimum of six days prior to the trials. For four weeks, the rats in each group were confined in wire cages and given the experimental food. The rats were divided into five primary groups, with six rats in each: First category, the normal or negative group (-Ve): Fed on basal diet only according to **Reeves *et al.*, (1993)**, being a negative control. Subjects in Group 2 who tested positive (+Ve) were given a high-fat diet (HFD) **Min *et al.*, (2004)** as a positive control. The third group received a high-fat diet (HFD) that included ground Purslane seeds at a rate of 5% per kilogram diet. The fourth group received a high-fat diet (HFD) that included 10% ground Purslane seeds per kilogram diet . The fifth group received a high-fat diet (HFD) that included ground purslane seeds at a rate of fifteen percent per kilogram diet.

Induction of obesity:

Rats were fed on high fat diet **(HFD)** containing (saturated fat 19%, soybean oil 1% to provide essential fatty acids, sucrose 10%, casein 14%, cellulose 5%, vitamin mixture 1%, salt mixture 3.5%, choline chloride 0.25% and the remainder is corn starch) for four weeks to induce obesity in rats **(Min *et al.*, 2004)**.

Biological evaluation

Increasing weight in kilograms (BWG) and as a percentage (BWG%) **Chapman *et al.*, (1959)**, by means of the subsequent equations:

$$\text{Body weight gain (BWG \%)} = \frac{\text{Final weight - Initial weight}}{\text{Initial weight}} \times 100$$

Peritoneal fat pad:

The rats' peritoneal fat pads were removed, weighed, and preserved at -20°C in accordance with the procedures outlined in **Azain *et al.*, (2000).**

Biochemical analysis

Scarification was performed after rats had fasted overnight at the conclusion of the 4-week study period. Blood samples were taken from each rat and spun in a centrifuge to extract serum for biochemical examination. Activity levels of the plasma aminotransferases aspartate (AST) and alanine (ALT) measured by **Reitman and Frankel, (1957),** Alkaline phosphatase (ALP) was assayed in serum according to the method of **Roy, (1970)**, total cholesterol (TC) **Allain., (1974)**, VLDL-c and triglycerides were determined in milligrams per deciliter **Fassati and Prencipe, (1982),** high density lipoprotein cholesterol (HDL-c) **Lopez, (1977)**, LDL-c, or low density lipoprotein cholesterol, was determined in milligrams per deciliter **Fridewald *et al.*, (1972)**. Uric acid according to the method of **Milena., (2003),** Urea and creatinine using the procedure outlined in **Young, (2001).** Glucose according to **Trinder, (1969),** leptin according to the methods described by **Zhang *et al.*, (1995).**

Statistical analysis

The mean ±standard error (SE) was used to display the collected data. To find the statistically significant differences between the various groups, we utilized an Analysis of Variance (ANOVA) test based on **Armitage and Berry, (1987)**. If the p-values were less than or equal to 0.05, we deemed all differences significant.

Result & discussion

According to table (1), rats given a high-fat diet had a 12.99% higher mean body weight than the negative control group ($P<0.05$). On the other hand, rats given purslane powder had a significantly lower mean body weight than the positive group ($P<0.05$). When compared to treatment

groups, rats given a food supplemented with 15% purslane powder (mean value was 3.15%), followed 10% and 5% (mean values were 5.15% and 8.75%,respectively), with significant different reduction. Additionally, when contrasted with the negative control group, the positive control group had considerably higher levels of peritoneal fat ($P<0.05$). There was a substantial decrease ($P<0.05$) in peritoneal fat in all groups treated with purslane as compared to the positive control group. The most recent data on weight loss was consistent with **Ramadan *et al.* (2017)**, according to whom mice given Purslane extract lost significantly less weight than rats given a placebo. **Abdalla, (2010),** suggested that Purslane ethanolic extract's anti-obesity effects might be attributed to a reduction in hepatic fatty acid production and fat intake, as well as an increase in fatty acid breakdown associated to energy expenditure .

Table (1): Effect of different levels of Purslane seeds on body weight

Groups	Initial weight (g)	Final weight (g)	BWG%	Peritoneal fat %
	Mean±SE	Mean±SE	Mean±SE	Mean±SE
-Ve	233.4±1.91^{a}	250.20±1.71^{bc}	7.22±0.30^{c}	3.31±0.20^{b}
+Ve	233.4±1.21^{a}	264.2±1.20^{a}	12.99±0.60^{a}	7.08±0.13^{a}
Purslane seeds 5%	235.6±1.24^{a}	256.4±1.69^{b}	8.75±0.20^{b}	2.96±0.10^{bc}
Purslane seeds 10%	238.4±1.20^{a}	251.0±1.30^{bc}	5.15±0.21^{d}	2.66±0.07^{c}
Purslane seeds 15%	239.2±1.77^{a}	246.6±2.01^{c}	3.15±0.20^{e}	1.96±0.11^{d}

Mean with different letters (a, b, c, d and e) in the same column differ significantly at ($P<0.05$).

There was a notable rise ($P<0.05$) in blood leptin hormone levels as compared to the negative control group, according to the data in table (2). Serum leptin hormone levels were lower in the Purslane powder seeds-fed rats compared to the positive control group. Serum leptin hormone levels were significantly lower in the rat group given a diet supplemented with 15% purslane compared to the 5% and 10% *Portulaca* powder seeds groups. In contrast to the negative control group, the mean glucose level was 70.4

mg/dL, the positive control group had a much higher level (112.4). The results demonstrated that rats given diets containing 5%, 10%, or 15% *portulaca* powder seeds had lower mean values (95.6, 83.4, and 80.00 mg/dL, respectively) as compared to the positive control group. The results were agreed with **Bendong, (2012),** discovered that the leptin hormone in overweight rats was decreased by *Portulaca oleracea*. **Mohamed, (2011),** investigated the impact of Purslane seeds on glucose levels. It demonstrated a substantial decrease in insulin, body weight, and blood glucose levels in the serum .

Table 2: Effect of different levels of Purslane seeds on serum glucose and leptin hormone

Groups	Glucose (mg/dL)	Leptin (µg/L)
	Mean±SE	**Mean±SE**
-Ve	70.4 ± 0.71^{d}	1.79 ± 0.09^{d}
+Ve	112.4 ± 1.69^{a}	5.14 ± 0.21^{a}
Purslane seeds 5%	95.6 ± 1.16^{b}	4.06 ± 0.14^{b}
Purslane seeds 10%	83.4 ± 1.61^{c}	3.45 ± 0.20^{bc}
Purslane seeds 15%	80.0 ± 1.01^{c}	2.83 ± 0.10^{c}

Mean with different letters (a, b, c, d and e) in the same column differ significantly at ($P<0.05$).

The results from table (3) showed that the average values of AST, ALT, and ALP were significantly higher ($P<0.05$) than in the negative control group. In contrast, as compared to the positive control group, rats given varying amounts of purslane powder (5%, 10%, and 15%) had lower levels of AST, ALT, and ALP, respectively. Findings corroborated previous reports that **Anusha *et al.*, (2011),** who conducted the Purslane experiment on rats who were given a high-fat diet. In addition to improving liver functions, *portulaca oleracea* decreased levels of hypoglycemia and hypolipidemic liver enzymes and had an antioxidant impact. Also, **Mohamed, (2011),** discovered that gamma glutamyl transaminase (GGT), liver alanine aspartate (ALT), and purslane seed powder led to a decrease in

serum GGT levels. **Abd El-Aziz *et al.* (2014),** showed that when compared to a positive control (injected with CCl4), there was a highly significant drop in serum AST, ALT, and ALP after administering Purslane aqueous extract, whether with or without CCl4. And yet., **Ahmida, (2010),** found that the phytochemicals included in Purslane, which included omega-3 fatty acids, β-carotene, flavonoids, and alkaloids, were responsible for its hepatoprotective effects .

Table (3): Effect of different levels of Purslane seeds on liver enzymes

Groups	AST (µg/L)	ALT (µg/L)	ALP (µg/L)
	Mean±SE	Mean±SE	Mean±SE
-Ve	57.0±0.70^{e}	22.40±2.90^{d}	50.51±1.99^{c}
+Ve	92.2±1.28^{a}	37.60±0.60^{a}	80.16±2.14^{a}
Purslane seeds 5%	84.8±1.20^{b}	31.80±0.40^{b}	70.19±2.95^{b}
Purslane seeds 10%	80.6±0.50^{c}	26.40±0.90^{c}	65.63±2.11^{b}
Purslane seeds 15%	69.80±0.90^{d}	24.6.00±0.57^{cd}	59.65±2.15^{b}

Mean with different letters (a, b, c, d and e) in the same column differ significantly at (P<0.05).

According to the data in table (4), there was a substantial rise (P<0.05) in T.C. and TG in the positive control group as compared to the negative control group. The data demonstrate that as compared to the positive group, the groups given varying amounts of Purslane (5%, 10%, and 15%) had significantly lower levels of total cholesterol and triglyceride (P<0.05). A extremely substantial reduction was observed in rats whose diets were supplemented with 10% and 15% purslane compared to the treated groups that received 5%. Rats given diets containing 5%, 10%, and 15% Purslane, respectively, had higher HDL-c and significantly lower LDL-c and VLDL-c levels compared to the positive control group, according to the data. Powdered purslane seeds reduced serum levels of T.G, T.C., and LDL-c while increasing HDL-c, according to another study that corroborated the present one **(Mohamed, 2011)**. Also, **Zidan *et al.* (2014),** showed that the hypercholesterolemia group had higher serum T.C. and

T.G. levels than the group that received Purslane extract. The current findings are consistent with **Ali *et al.* (2014),** who noted that there were statistically significant changes over time in the Purslane with respect to T.C, LDL-c, and T.G ($P<0.05$). Only LDL-c and T.G. showed statistically significant changes in the indicated parameters; the others showed no such difference. **Niharika and Sukumar, (2016)**, demonstrated that Purslane extract increased HDL-c while decreasing T.G, T.C, LDL-c, and VLDL-c .

Table (4): Effect of different levels of Purslane seeds on lipids profile

Groups	TC (mg/dL)	TG (mg/dL)	HDL-c (mg/dL)	LDL-c (mg/dL)	VLDL-c (mg/dL)
	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
-Ve	89.1±0.84^{e}	77.3±3.2^{d}	57.3±0.60^{a}	16.3±0.9^{d}	15.46±0.16^{d}
+Ve	145.4±0.84^{a}	152.2±9.0^{a}	40.3±0.59^{d}	74.7±1.1^{a}	30.44±0.17^{a}
Purslane seeds 5%	111.7±1.27^{b}	114.8±6.1^{bc}	44.96±0.57^{c}	43.8±0.8^{b}	22.96±0.24^{c}
Purslane seeds 10%	98.8±0.89^{c}	117.0±6.1^{bc}	48.5±0.46^{b}	26.9±0.5^{c}	23.4±0.16^{c}
Purslane seeds 15%	100.3±0.83^{cd}	126.2±5.2^{b}	50.6±0.60^{b}	24.5±1.5^{c}	25.24±0.17^{bc}

Mean with different letters (a, b, c, d and e) in the same column differ significantly at ($P<0.05$).

VLDL= TG/5

TC= Hdl+Ldl+Vldl

Table 5 shows that rats given varying amounts of Purslane (5%, 10%, and 15%) had considerably lower levels of urea, uric acid, and creatinine compared to the positive control group, with a significance level of $P<0.05$ for each. **Karimi, (2010),** demonstrate that Purslane extract may protect from renal toxicity and prevent renal injury, since therapy with extracts of the plant decreased blood urea nitrogen (BUN) and serum creatinine levels without causing any harm to the tubules. **Shirwaikar *et al.* (2003),** Animals given Purslane had lower uric acid levels, which could be a result of the plant's antioxidant properties.

Table (5): Effect of different levels of Purslane seeds on serum urea, uric acid, and creatinine

Groups	Urea (mg/dl)	Uric acid (mg/dl)	Creatinine (mg/dl)
	Mean±SE	Mean±SE	Mean±SE
-Ve	25.0±0.40^{d}	3.40±0.30^{c}	0.51±0.01^{c}
+Ve	62.2±1.88^{a}	6.36±0.17^{a}	2.23±0.09^{a}
Purslane seeds 5%	56.8±0.60^{b}	5.26±0.20^{b}	0.76±0.01^{b}
Purslane seeds 10%	47.6±0.50^{c}	4.79±0.10^{b}	0.71±0.01^{bc}
Purslane seeds 15%	42.4±070^{c}	3.89±0.17^{c}	0.66±0.01^{bc}

Mean with different letters (a, b, c, d and e) in the same column differ significantly at ($P<0.05$).

Conclusion:

Different levels of Purslane powder may improve body weight, lipid profile, liver enzymes, and kidney functions. They will be potentially suitable for use for obesity patients.

References

- Abd El-Aziz, A., Sobhy M., Ahmed, K., Abd El Hameed, A., Rahman, Z. and Hassan, W. (2014): Chemical and remedial effects of purslane (Portulaca oleracea) plant. Life Science Journal, 11 (6): 31-42.
- Abdalla, H. (2010): Purslane extract effects on obesity-induced diabetic rats fed a high-fat diet. Mal. J. Nut., 16 (3): 419-429.
- Ahmida, M. (2010): Evaluation of in vivo antioxidant and hepato-protective activity of Portulaca oleracea L. against paracetamol induced liver toxicity in male rats. Am. J. Pharmagol., 5: 167-176.
- AL-Faris, N.A. (2014): Biological evaluation of the weight reduction capacity of selected herbal products available in the Kingdom of Saudi Arabia. Nutrition and Food Sciences J., 3 (1): pp. 7-13.
- Ali, M., Roya, K. and Hadi, J. (2014): The effect of Portulaca oleracea on obesity. Med Arch Jun, 68 (3): pp.198-199.

- Armitage, G.Y. and Berry, W.G. (1987): Statistical methods 7thEd. Ames., Iowa State University. pp 39-63.
- Anusha, M., Venkateswariu, M. and Prabhakaran, V. (2011): Hepatoprotective activity of aqueous extract of Portulaca oleracea in combination with lycopene in rats. Indian J Pharmacol., 43 (5): pp. 563–567.
- Azain, M., Hausman, D., Sisk, M., Flat, W. and Jewell, D. (2000): Dietary conjugated Linoleic acid reduces rat adipose tissue cell size rather than cell number. J. Nutr., 130: 1548-1554.
- Chapman D.G., Gastilla, R. and Campbell, J.A. (1959): Evaluation of protein in foods: 1- A Method for the determination of protein efficiency ratio. Can. J. Biochem. Phys; 37: pp. 679- 86.
- Fassati, P. and Prencipe, L. (1982): Determination of serum triglycerides: Clin. Chem.; 28 (10): 2077- 2080.
- Fridewald, W., Leve, R. and Fredrickson, D. (1972): Estimation of the concentration of low-density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. Clin Chem, 18: pp. 499-502.
- International Association for the Study of Obesity (IASO) (2012): [Graphic illustration using a world map] Global prevalence of obesity in adult females.
- Karimi, G., Khoei, A., Omidi, A., Kalantari, M., Babaei, J. and Taghiabadi, E. (2010): Protective effect of aqueous and ethanolic extracts of Portulaca oleracea against cisplatin induced nephron-toxicity. Iranian Journal of Basic Medical Sciences, 13 (2). pp. 31-35.
- Lopez, M.F. (1977): Determination of serum high density lipoprotein cholesterol: Clin. Chem; (23): 882.
- Min, L., Ling, S., Yin, L., Stephen, C., Randy, J., David, D. and Patrick, T. (2004): Obesity induced by a high fat diet down regulates apoliprotein A-IV gene expression in rat hypothalamus. Am J physiol Endocrinol Metab, 287: pp. 366-370.
- Milena, J. (2003): Predrag Djurdjevic and DejanStankov .JSCS. 68(8-9): 691-698.
- Mohamed, K. (2011): Effect of portulaca oleracea seed in treatment of type-2diabetes mellitus pation as adjunactive and altractive theraps.journal of Ethanopharmacology, 137: pp643-651

- .Niharika, S. and Sukmar, D. (2016): Hypolipidemic effect of Portulaca oleracea in rats fed on high cholesterol diet. J nutrition and food science, 30: pp.120-125.
- Ramadan, B., Schaalan, M. and Tolba, A. (2017): Hypoglycemic and pancreatic protective effects of Portulaca oleracea extract in alloxan induced diabetic rats. Complementary and Alternative Medicine, 17 (37): 1-10.
- Reeves, P., Nielsen, F. and Fahey, G. (1993): AIN-93 purified diets for laboratory rodents. Final report of the American Institute of Nutrition of the AIN -76A rodent diet. J Nutr,51: pp. 123-1951.
- Reitman, A. and Frankel, S. (1957): Determination of serum glutamate oxaloacetate transaminase (GOT) and serum glutamate pyruvate transaminase (GPT): Am. J. Clin. Path., 28: 56.
- Roy, S. (1970): Colorimetric method of serum alkaline phosphatase. Journal of Clinical Chemistry, 16: 431-432.
- Shirwaikar, A., Malini, S. and Kumari, S. (2003): Protective effect of Pangamia pinnata flowers against cisplatin and gentamicin induced nephrotoxicity in rats. Indian J. Exp. Biol., 41: 58-60.
- Trinder, P. (1969): Determination of blood glucose using 4-amino-phenazone. J. Clin. Path., 22: 246.
- World Health Organization (WHO) (2010): World Health Statistics 2010. Geneva, Switzerland: World Health Organization.
- Young, D.S. (2001): Effects of disease on clinical Lab. tests, AACC. Clinical Chem, 48, 682.
- Zidan, Y., Bouderbala, S., Djellouli, F., Lacaille-Dubois, M. and Bouchenak, M. (2014): Portulaca oleracea reduces triglyceridemia, cholesterolemia, and improves lecithin: cholesterol acyltransferase activity in rats fed enriched-cholesterol diet. Phytomedicine, 21(12): 1504-1508.(
- Zhang, Y., Proenca, R., Maffei, M., Barone, M., Leopold, L. and Friedman J. (1995): Positional cloning of the mouse obese gene and its human homologue. Nature; 372 (6505): 425–432.

تقييم تأثير المستويات المختلفة من الرّجلة على مكافحة السمنة لدى الجرذان البيضاء

د. هاني حمدي الجزار سمر شعوير ناصر نسيم عطية زهران

الملخص العربي:

السمنة من المشاكل الصحية التي يعاني منها العالم، فهي تؤدي إلى زيادة تراكم الدهون وزيادة الوزن في الجسم. والهدف الرئيسي من هذه الدراسة هو معرفة تأثير بذور الرجلة في مكافحة السمنة. وقد تم تقسيم ثلاثين من ذكور الجرذان البيضاء البالغة إلى خمس مجموعات رئيسية (٦ جرذان لكل مجموعة) على النحو التالي: المجموعة (١) المجموعة السلبية أو الطبيعية (Ve_-): تم تغذيتها على النظام الغذائي الأساسي فقط كمجموعة تحكم سلبية. المجموعة (٢) المجموعة الإيجابية (Ve_+): تم تغذيتها على نظام غذائي عالي الدهون (HFD) كمجموعة ضابطة إيجابية. المجموعة (٣): تم تغذيتها على نظام غذائي عالي الدهون مضافاً إليه بذور الرجلة المطحونة بنسبة ٥٪. المجموعة (٤): تم تغذيتها على نظام غذائي عالي الدهون مضافاً إليه بذور الرجلة المطحونة بنسبة ١٠٪. المجموعة (٥): تم تغذيتها على نظام غذائي عالي الدهون مضافاً إليه بذور الرجلة المطحونة بنسبة ١٥٪. وقد أشارت النتائج إلى أن إضافة الرجلة أدت إلى انخفاض معنوي ($P<0.05$) في وزن الجسم للجرذان البدينة مقارنة بالمجموعة الضابطة الإيجابية. علاوة على ذلك، انخفضت مستويات الجلوكوز في المصل، واللبتين، والكوليسترول، والدهون الثلاثية، و$LDL-c$، و$VLDL-c$، واليوريا، وحمض البوليك، والكرياتينين، وAST، وALT، وALP بشكل ملحوظ، في حين ارتفعت مستويات $HDL-c$ في المصل بشكل ملحوظ.

الكلمات الدالة: بذور الرجلة، وزن الجسم، دهون الدم، وظائف الكلى، إنزيمات الكبد