



Study the Sub acute Effect of *Eruca sativa* Leaves Extract on Hematological Parameters of Male Rats

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Abstract

Eruca Sativa is a plant usually used as antioxidant, anti-inflammatory and used for salad preparations and in the traditional medicine. This study aimed to determine the effect of crude ethanolic extract of *Eruca Sativa* leaves on the hematological changes. Twenty albino male rats were divided for four groups, group D, rats served as control. Group A, B, and C, rats dosed orally 200, 400 and 600 mg/kg respectively for one week. The results indicate that white blood cell and the Red blood cells were significantly increased ($p < 0.05$), while HCT, RDW-SD, RDW-CV, PLT, MPV and PCT was significant ($p < 0.05$) changes. On the other hand the HGB, MCV, MCH, MCHC and PDW showed no significant ($p < 0.05$) effects.

Key Words: hematology, blood and plants.

الخلاصة

نبات الجرجير من النباتات التي تستخدم كمضاد للاكسدة، مضاد للالتهابات ويستخدم في تحضير السلطة و في الطب التقليدي. الهدف من الدراسة هو تحديد تأثير مستخلص اوراق الجرجير على التغيرات الدمية. قسمت عشرون من الجرذان نوع البينو الذكور الى اربع مجاميع. المجموعة الاولى استخدمت كمجموعة سيطرة، المجموعة الثانية والثالثة والرابعة جرعت عن طريق الفم بجرعة 200 , 400 و 600 ملغم/كغم على التوالي لمدة أسبوع. اظهرت النتائج وجود زيادة معنوية على مستوى ($p < 0.05$) في كريات الدم البيض والحمر. وبينما هنالك تغيرات معنوية على مستوى ($p < 0.05$) في كل من المعايير HCT, RDW-SD, RDW-CV, PLT, MPV and PCT , بالجانبا الآخر لا توجد أي تغيرات معنوية في المعايير HGB, MCV, MCH, MCHC and PDW بمستوى معنوية ($p < 0.05$).

Introduction

Some medicinal herbs and seeds have been well known since the old civilization of ancient Egyptians, Chinese and Greek. The most of the antibiotic growth promoters has been embargo in more countries, particularly in USA and the European Union, because remain their residues in the animal products, with the development of bacterial resistance to antibiotics [1] and [2]. Therefore, search for alternative safe growth promoters is urgent. The new trend in rat production is using of many medicinal seeds plants in

rabbits and immune response of rabbits ration to enhance productive performance.

Rocket seed meal (*Eruca sativa*) locally known as jarjeer contain vitamin C, carotenoids, , flavonoids such as luteolin, apin and glucosinolates the precursors of sulfaraphene and isothiocyanates [3] also contain volatile oils like apiola , -B-phellandrene and myristicin [4] [5]. Glucosinolates posses several biological activities including antioxidant action, anti-carcinogenic, anti-fungal, and anti-bacterial

[6]. They also contain Zinc, Copper, Iron, Magnesium, Manganese and other elements [7] that it increase immune response with enhance the reproductive performance carotenoids that it can guard phagocytic cell from antioxidative damage. Enhancement B and T lymphocyte proliferative responses and elevate the production of certain interleukins [8]. As we as, they increase plasma IgG level [9]. It's known as anti-inflammatory, diuretic and affects on content blood circulation. *Eruca* seeds have high level of oil, erucic acid and protein glucosinolate contents and commonly used as diet in animal feed in region of Asia, particularly in Pakistan and India [10].

Materials and methods

Plant material and preparation of extract:

Fresh *Eruca sativa* leaves were purchased from Najaf, a local vegetable market in Iraq, and the identity of these leaves were confirmed by Department of Seed Diagnosis and Certification dependent to the Ministry of Iraqi Agriculture. After identification of *Eruca sativa*, the leaf part of the plant was cleaned, dried in the shaded place for 30 days then were finely powdered by electrical blender. The powdered leaves were stored in dark glass to protect them from light and moldiness. The amount of 100 g of rocket leaves were placed in a glass percolator with 1000 ml of ethanol and were admitted to stand at room temperature for about 72 h, the mixture, after 3 days it was filtered by a fine muslin cloth followed by specific filter paper (Whatman No. 1). The mixture was then distilled under evacuated pressure in a rotary evaporator (RE 200B/UK) [11]. The plant extract was then it dissolved in Dimethyl Sulfoxide (DMSO) and administered orally to rats with different doses [12].

Experimental animals: In this experiment twenty healthy adult male *Rattus norvegicus*

rats were obtained from Animal House, Faculty of Veterinary Medicine/ Kufa University. Twenty male rats (6-8 weeks old) weighed between 150g – 200g were kept at room temperature in normal humidity rooms on a standard dark/light cycle (12 h dark; 12 h light cycle) at (25±2C°). Each 5 rat was placed in separate plastic cages (56 × 39 × 19) covered the cage with wooden chips in the animal house. The rats were fed with standard rat diet food of pellets and tap water [13]. They were kept under observation for about 15 days of adaptation before the initiation of the experiment. All the procedures described were showed and approved by Institutional Animal Ethical Committee. During the experiments, all animals got human care applying to the criteria outlined in the “Guide for the Care and Use of laboratory Animals” prepared by published by the national Institute of health and the National Academy of Sciences.

Twenty male rat were divided into four group(five for each), Group A given orally 200 mg/kg body weight ES extract, Group B given orally 400 mg/kg body weight ES extract, Group C given orally 600 mg/kg body weight ES extract, All groups given extract daily until the end of experiment, while group D leave untreated as a control group.

Behavioral observation: During the study period, the oversight of behavioral signs was observed daily for all groups of rats. Detad observations of the individual rats were made daily in comparison with the normal group rats. Observations included vision evaluations of the skin for any sign of respiration (dyspnoea), exophthalmia, convulsion, salivation, ,and any changes in movement such as whether the animals tend to actively moving or stay peaceful in their cage.

Blood collection: Blood samples were taken from the rats through cardiac puncture then put in plane tubes without ethylene diamine tetraacetic acid (EDTA), kept at room temperature 25° for (20-30) minute, centrifuge for [14] minute at (3000) rpm for 20 minutes [15].

Table (1). The effect of *Eruca sativa* leaves extract on white blood cells and red blood cells of Rat

parameters groups	WBCs 10^9 /L	RBCs 10^{12} /L
Control	11.33± 1.78 b	7.94 ± 0.18 b
200mg/kg	8.94 ± 1.19 b	7.87 ± 0.29 b
400mg/kg	14.32± 2.94 a	7.41 ± 0.19 ab
600mg/kg	6.84± 2.01 ab	8.18 ± 0.30 a

Differences in the same column with different superscripts are statistically significant at $P < 0.05$.

The effect of *Eruca sativa* on WBCs and RBCs in all groups are presented in this table, the results regarding WBCs and RBCs appeared that there was significant increase ($p < 0.05$) with 400 mg/kg and 600 mg/kg compared with the control group. Concerning the total white blood cells, rat given 400 mg/kg and 600 mg/kg induced significant ($p < 0.05$) increase in WBCs (14.32 ± 2.94 and 6.84 ± 2.01) respectively compared with the other two groups, this may be due to high contents of indirect and direct antioxidants [16] [17]. While the

Statistical Analysis

Data analysis was done by the Graph Pad prism computer software. one-way analysis of variance (ANOVA) were used for comparison and Students 't'-test. A P-value < 0.05 was considered significant.

Results and discussion:

improvement of RBCs in 400 mg/kg and 600 mg/kg (7.41 ± 0.19 and 8.18 ± 0.30) respectively may be due to increase elevate of immunity and reduce the negative effect of aflatoxin B1 due to active ingredients of *Eruca sativa* [18]. This agree with the resulting of [19] who reported that the addition of dried Rocket (*Eruca sativa*) to the fish diets has improved the hematological blood and disagree with [20] who reported that the addition of rocket (*Eruca sativa*) to the diet of rabbit male didn't alter the hematological blood.

Table (2): The effect of *Eruca sativa* leaves extract on hemoglobin and hematocrit of Rat

parameters groups	HGB g/dL	HCT %
Control	14.1± 0.20	42.7 ± 0.82 a
200mg/kg	13.7± 0.50	40.9 ± 1.09 b
400mg/kg	13.5± 0.49	40.2 ± 1.03 b
600mg/kg	14.5± 0.60	42.3 ± 1.17 ab

Differences in the same column with different superscripts are statistically significant at $P < 0.05$.

Table (2) shows no significant effect ($p < 0.05$) of 200, 400, and 600mg/kg respectively on the hemoglobin and hematocrit. This agree with [21] who reported that the added of radish and rocket cakes to Hubbard broiler chicks diets didn't change the hemoglobin percentage, which

indicated that there was no anemic occurrence and that it was safer to use these plants in animals diets. On the other hand hematocrit show significant decrease ($p < 0.05$) for the 200 mg/kg and 400 mg/kg compared to the control group (40.9 ± 1.09 and 40.2 ± 1.03). this agree with [20] who found that the rocket decrease significantly of hematocrit HCT after use it in rabbit diets. This may be due to adverse effect of high level protein of *Eruca sativa* on RBCs volume.

Table (3): The effects of *Eruca sativa* leaves extract on, mean corpuscular hemoglobin concentration (MCHC), and mean corpuscular hemoglobin (MCH) and on the mean corpuscular volume (MCV).

parameters groups	MCV fL	MCH pg	MCHC g/L
Control	53.9 ± 1.32	17.8 ± 0.26	331 ± 3.80
200mg/kg	52.0 ± 1.10	17.6 ± 0.46	335.6 ± 2.56
400mg/kg	54.1 ± 0.75	18.2 ± 0.23	335.2 ± 3.12
600mg/kg	52.2 ± 0.85	17.5 ± 0.31	334.4 ± 3.52

Differences in the same column with different superscripts are statistically significant at $P < 0.05$.

In table 3 MCV, MCH and MCHC respectively had no significant. This may be due to MCV, MCH and MCHC weren't affected by 200, 400 and 600 mg/kg. The obtained results are in accordance with those reported by [22] they found that MCV, MCH and MCHC weren't affected by feeding (*Nigella sativa*) to Hammary $\times$ Dorset male lambs. This agree with [23] they found there weren't significant effect ($p < 0.05$) in MCH, MCV and MCHC to some medicinal plants like (*Nigella sativa*), (*Trigon-ella foenum*) and (*Eruca Sativa*) of male Awassi lambs . This is may be due to the dose of *Eruca Sativa* at 200, 400 and 600mg/kg not affected the blood of rats or the time of experiment must be longer to show significant effect.

Table (4): The effect of *Eruca Sativa* leaves extract on Red Blood Cell Distribution Width Standard Deviation (RDW-SD) and Red Blood Cell Distribution Width Coefficient of Variation (RDW-CV)

parameters groups	RDW-CV %	RDW-SD fL
Control	0.199 ± 0.08 a	46.7 ± 3.10 a
200mg/kg	0.191 ± 0.09 ab	43.0 ± 1.53 ab
400mg/kg	0.185 ± 0.06 b	43.5 ± 2.11 b
600mg/kg	0.173 ± 0.04 b	39.6 ± 1.14 b

Differences in the same column with different superscripts are statistically significant at $P < 0.05$.

The study show significant change ($p < 0.05$) in RDW-CV and RDW-SD. This study showed clearly that *Eruca Sativa* extract does possess hematopoietic activity and is not any hematotoxic. This results disagree with [24] they found that the methanolic leaf of *Jatropha curcas* didn't affect the RDW-CV and RDW-SD in male albino rats. Biochemical and hematological incidences have been reported to be a reliable parameter for assessment of the health situation of animals [25] [26].

Table (5): The effect of *Eruca Sativa* leaves extract on the Platelet count (PLT), Platelet Distribution Width (PDW), Mean Platelet Volume (MPV), and Plateletcrit (PCT).

parameters groups	PLT 10 ⁹ /L	MPV fL	PDW mL/L	PCT mL/L
Control	970 ± 51.3 ab	8.8 ± 0.16 ab	15.4 ± 0.06	7.88 ± 0.27 b
200mg/kg	1076 ± 68.4 a	7.4 ± 0.12 a	15.4 ± 0.09	8.85 ± 0.44 a
400mg/kg	935.2 ± 41.1b	6.8 ± 0.16 b	15.3 ± 0.08	6.47 ± 0.18 ab
600mg/kg	950.2 ± 36.4 b	6.8 ± 0.08 b	15.4 ± 0.06	6.57 ± 0.70 b

Differences in the same column with different superscripts are statistically significant at $P < 0.05$.

Table 6 show no significant ($p < 0.05$) effect in PDW, on the other hand show significant ($p < 0.05$) increase in MPV and PCT in the 200mg/kg (7.4 ± 0.12 and 8.85 ± 0.44) respectively compared with control and other treatments. And there is significant decrease ($p < 0.05$) at the 400 and 600 mg/k respectively in platelet count. The increase in blood platelet count (thrombocytopenia) may decrease the coagulating factors that produced by platelets. This may be one or more contents of rocket that affected on bone marrow into produce more platelets in the blood stream.

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