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# Assessment of Toxicological Effect of *Lactuca serriola* L. in Sprague-Dawley Rat

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**Abstract**

THIS STUDY aimed to assess the toxicity level of the alcoholic extract of *Lactuca serriole. L.* A total of thirty mature male rats were used. The animals were categorized into five distinct groups: Group 1, referred to as the control negative group, did not receive any treatment. Groups 2, 3, 4, and 5, known as the treatment groups, were given different dosages of *Lactuca* extract through gavage. The dosages administered were 100, 200, 400, and 800 mg/kg/bw, respectively. This treatment was carried out for 14 days. Cardiac blood samples were collected, followed by biochemical testing. Tissue sections from the kidney, liver, and spleen were prepared for H&E staining. There was no notable disparity in body weight between the treated groups and the control group. However, there was a substantial enhancement in the reduction of white blood cells (WBCs) and lymphocytes in the groups that received a maximum dosage of *Lactuca*. In addition, *Lactuca* significantly increased the thrombocyte differential count, all blood lipid indicators, and liver enzyme markers compared to the control group. The rats who received doses of 400 and 800 mg/kg/bw exhibited significant histopathological damage in their liver, kidney, and spleen. In contrast, the rats given doses of 100 and 200 mg/kg/bw had normal organ structures. These findings may provide new and valuable information about the safe dosage range (100-400) of phenolic compounds from Lactuca that can be used in experiments with animals.

**Keywords:** *Fatty degeneration, Lactuca serriola, Liver enzyme, Lymphocytic hyperplasia, Sharbazher district.*

## Introduction

The potential impact of eating a variety of natural food resources of vegetables, fruits, and grains has been revealed in many studies in the prevention, postponement, and progression of many illnesses. Designating this practice for the benefit of humanity is very crucial. To help the well-being of human development. Utilizing essential culinary herbs as a preventive measure against the development of specific illnesses is highly valued. This has led to a surge in research to discover another form of treatment, such as herbal remedies [1-3]. In comparison with manufactured drugs, natural remedies showed remarkable safety profiles and better outcomes [4, 5].

It has been experienced that numerous medicinal plants provide effective drugs that are readily available, with reasonable prices, and hence beneficial for treating specific diseases [6]. A necessary section of healthcare includes the recognition of plants that possess physiologically active chemicals. In accordance with the World Health Organisation reports, 60% of the global population relies on herbal remedies, whereas over

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80% of individuals in underdeveloped countries rely almost entirely on these remedies for their fundamental healthcare needs. Phytochemicals and their chemical analogs have yielded abundant medically beneficial drugs for the management of both long-term and sudden illnesses [7].

*Lactuca serriola* L. (*L. serriola*) belongs to the *Asteraceae* family and is well-recognized for its therapeutic properties [8]. This herb is thought originally domesticated from the wild type of *L. Serriolais* in the Mediterranean regions and Southeast Asia. The plant is a herbaceous plant that has an almost life span of one or two years. The plant stems are tall, and spherical that is covered with prickles, and light green. The object displays a limited number of toothed edges and possesses leaves that are arranged alternately and divided into lobes along a central axis [9]. The chemical screening tests for the existence of bioactive compounds in the plant established the availability of various substances, including lactucin, lactucone, lactucic acids, lactucopicrin, sesquiterpene esters, beta-carotene, vitamins, oxalic acid, iron, alkaloids, and phenolic groups. Additionally, the accessibility of many sedative mediators, hypnotic, expectorative agents, coughing suppressant substances, vaso-relaxant, diuretic, purgative, qualities demulcent, antispasmodic, and antibacterial ingredients has recognized the plant as an effective traditional remedy against diverse health issues [10].

Furthermore, there have been reports of the conventional use of *Lactuca serriola* L. for treating ophthalmological conditions, bronchitis, cough, headaches, and various other diseases [11]. A pharmacological study was conducted to investigate the sedative, antioxidant, hepatoprotective, anti-inflammatory, and anti-carcinogenic characteristics of the substance [12].

We are inspired to conduct this study to evaluate the toxicity level of this plant before using it for various therapeutic uses, due to its significance as a medication and its richness of antioxidants.

## Material and Methods

### *Collection of plant*

The specimens were gathered in the Sharbazher district of Sulaimani, Iraq, during March and April in the year 2022. The plant samples were validated by the Botany Department at the College of Agriculture, University of Sulaimani, Iraq**.**

### *Preparation of extract*

Sharbazher district experiences the flourishing of *Lactuca serriola* L in the spring season. The upper ground part of the plant was collected, including the leaves and stems. The herb part was fragmented and dehydrated at ambient temperature. The desiccated portions of *Lactuca serriola* L. The *serriola* was grinded and then stored in an airtight glass bottle. The preliminary sample for analysis had a combined weight of 350 grams. The dried powder of the plant was extracted using the maceration extraction process for two weeks. This involved soaking the raw material in a solvent consisting of 70% aqueous methanol. The mixture was then passed through gauze cloth and placed in an Amber reagent container, which was kept refrigerated for a further two weeks. After repeating the operation not less than three times, the filtrate which was a transparent solution was put in the refrigerator. The solvent was separated from the mixture by using a rotary evaporator. Finally, the solution was lyophilized by freeze dryer.

### *Animal Model and Experimental Design*

A total of thirty male rats, with weights ranging from 100 to 150 g, were acquired from the Teaching Hospital Animal House of the College of Veterinary Medicine, University of Sulaimani in Sulaimani, Iraq. The animals were monitored in a controlled environment setting with regulated temperature and lighting conditions. They were provided unrestricted access to tap water and a standard diet. The experimental structure was approved by the Ethical Committee of the College of Veterinary Medicine at the University of Sulaimani, which was done in compliance with the Guide for the Care and Use of Laboratory Animals (No: 030516). Five cohorts of rats were established. Before acquiring the diverse L. For the administration of *L. serriola* extract doses, participants were required to undergo a complete overnight fast without consuming any liquids. The groups (G2 to G5) were treated with different dosages of the extract (100, 200, 400, and 800 mg/kg/bw) through gavage feeding. In contrast, the negative control group G1 (n=6) received normal saline as a control. The extracts were dissolved in distilled water. The treatment was orally provided to the rats in groups 2, 3, and 4 for 14 days, which included two consecutive days of treatment with a one-day gap in between.

### *Clinical observations and body weight measurements*

Throughout the 14-day experimental period, the animals were monitored twice a day for signs of acute poisoning, including tremors, drowsiness, unsteady movements, seizures, unconsciousness, muscle contractions, diarrhea, itching, bent tails, hair loss, death, and any other odd behaviors. During the experiment, the researchers recorded the body weights of the rats on three occasions**:** at the beginning, post one week, and post two weeks (days 0, 7, and 14).

### *Blood sample collection and analysis*

On the 14th day of the experiment of sample collection, the animals were visually observed to confirm that the animals were safe and there were

unexpected signs and symptoms. Thereafter, the animals were anesthetized by using Ketamine and Xylazine. Through a cardiac puncture, 5 mL of blood was drawn, subsequently, 2 mL of blood was added into an EDTA blood collection tube and the rest of the blood was injected into a Gel and Clot activator. The Blood sample used for complete blood count (CBC) and the serum collection tube were incubated at normal room temperature for almost (20 to 30 minutes), after that the tubes were centrifuged at 3000 rpm for 15 minutes to yield sufficient serum. Finally, several chemistry tests were carried out by using Roche Cobas c111 analyzer including, (serum alkaline phosphatase (ALP), serum alanine aminotransferase (ALT), serum aspartate aminotransferase (AST), serum total proteins, serum albumin, serum bilirubin, serum triglycerides (TG), serum cholesterol, low-density lipoproteins (LDL), and high-density lipoproteins (HDL).

*Tissue sampling and histopathological examination*

After scarifying the animals several organs were collected including, kidneys, liver, and spleen. The organs were then rinsed with normal saline, trimmed to the appropriate dimensions, and fixed in 10% formalin solution for 48 hours. Subsequently, the sections were exposed to several stages of tissue processing which were then affixed on glass slides and stained with eosin and hematoxylin dyes. Two pathologists, who were unaware of the experimental design, examined the pathological characteristics and evaluated the lesions using various magnifications of light microscopy (Amscope$^{TM}$, Japan) (Table 1).

*Statistical analysis*

GraphPad Prism 9 was used to find the statistical differences between the mean and standard deviation of the treated and untreated groups. One-way analysis of variance (ANOVA) was run. Then to compare the (mean± STD) of body weight comparison of the groups. A two-way analysis of variance (ANOVA) with a mixed model was used. A p-value less than 0.05 was considered to be statistically significant.

## Results

*Body weight measures and the effects of Lactuca serriola L. extract*

Throughout the experiment, no deaths were seen with no sign of poisoning in the animals. The body weight comparison between the mean and standard deviation of treated and control groups showed insignificant increases in G1, G2, and G4 from (day 7 to day 14) compared to their preliminary weights on (day 0). Additionally, any changes have not been noticed in G3 during the experiment. However, the statistical significance declined (P= 0.01) in G5 on days 7 and 14 in comparison to day 0 (Table 2 and Fig. 1).

*Effect of Lactuca serriola L. extract on blood cells*

Hematological tests are vital methods to indicate various biological and pathological situations. Complete blood counts (CBC) were performed for both control (G1) and experimental groups (G2, G3, G4, and G5) that were treated with different concentrations of *Lactuca serriola* L. extract (100, 200, 400, 800 mg/ml) respectively.

The differential leukocyte counts (Table 3 and Fig. 2A), statistical analysis revealed that the mean WBC value in G2 (14.35± 1.85) was significantly higher than that of G1 (9.23± 0.64), with a p-value of (0.01). When compared to the control group, the remaining groups (G3, G4, and G5) did not exhibit any appreciable differences

The lymphocyte count of the treated animals (Table 3 and Fig. 2A). The mean values of G3 (6.10±1.21), G4 (3.75±1.85), and G5 (4.15±0.35) decreased significantly ($P<0.05$) compared to the mean of control, whereas the G2 (8.95±0.35 showed no statistical difference ($p>0.05$).

The lymphocytes%; of treated groups (G2, G3, G4, and G5) were extremely declined compared to G1 (94.30±1.80). The p values for these groups were (0.0002, 0.0073, 0.0002, and 0.0008) separately.

There is a significant increase in MID count in G2 (2.95±0.65) and G3 (1.57±0.31), with p values of (<0.0001 and 0.0426) unlike the previous groups, no significant change shown in G4 (1.05±0.25) and G5 (1.00±0.30) in comparison to the untreated group (0.63± 0.25).

The MID cell percentage is significantly higher in treated groups than the untreated group, G2 (20.90±1.9), G3 (15.57±1.90), G4 (20.00±3.60), and G5 (16.70±2.01). The p values for these groups are (<0.0001, 0.0003, <0.0001, and 0.0002) correspondingly.

The GRA count is meaningfully increased in G2 (2.45±0.85) and G3 (1.37±0.40). The p-values for these groups are (0.0003 and 0.0236). Nevertheless, no significant change is noticed in the mean values of G4 (0.90±0.10) and G5 (0.95±0.35).

Regarding the GRA% as shown in (Table 3) dramatically elevated in all treated animals G2 (16.85±3.95), G3 (12.20±0.82), G4 (18.20±4.70), and G5 (16.35±2.85). The p-values for these groups are (0.0003, 0.0048, 0.0002, and 0.0004) respectively.

The differential erythrocyte counts are shown in (Table 4 and Fig. 2B) which include several parameters for the estimation of the RBCs ratio and its constituents which help to diagnose many health conditions. The mean value and standard deviations of RBCs in G5 (7.85± 0.87) is the only group with that statistically significant *P* value (0.0087). However, no significant change is observed in other groups (G2, G3, and G4) compared to the control group (10.52± 0.58).

In addition, the hemoglobin level of animals was noticed, the mean value of G5 (14.90± 1.60) was the only group that had declined hemoglobin level *P* value (0.0285). Nevertheless, no statistically significant change was observed in other treated groups related to the control.

Hematocrit level (HCT) is exhibited in (Table 4 and Fig. 2B), the mean value of the treated and untreated groups was statistically considered, G3 (42.07± 2.48) and G5 (43.85± 3.85) where the two groups' HCT level diminished (*P* value for these groups were 0.0024 and 0.006) correspondingly to the control group G1 (56.00± 2.76). On the other hand, there were no significant variations seen in these parameters such as (MCV (fL), MCH (pg), MCHC (g/Dl), RDWa (fl), and RDW (%).

The differential thrombocyte counts are exposed in (Table 5 and Fig. 2C) which contain numerous parameters for the assessment of PLT count. The mean value of platelets in all groups; G2 (553.50± 4.5), G3 (680.00± 3.61), G4 (672.33± 6.51), and G5 (400.33± 6.03) were significantly raised p-value for all treated groups were (<0.0001) compared to G1 (138.00± 5.57).

The Mean platelet volume (MPV) of G5 (6.34± 0.29) was the only group-raised p-value (0.0499) compared to the untreated rats G1 (7.43± 0.76).

The Platelet hematocrit (PCT%) was raised in G3 (0.43± 0.06) and G5 (0.53± 0.04) corresponded to the untreated group G1 (0.09± 0.02) that p-value for these groups were 0.0139 and 0.0028.

Platelet-large cell ratio (P-LCC) revealed that a significant rise is noticed in the mean value of the treated animals G2 (22.52± 3.75), G3 (63.93± 8.10), G4 (50.27± 3.37), and G5 (42.90± 1.35) compared to the untreated ones G1 (11.00± 2.00). (p-value were 0.0299, <0.0001, <0.0001, and <0.0001) correspondingly.

In contrast, there were no significant statistical differences observed in these parameters (PDWa (fL) PDW (%), and P- LCR (%)) between the control and treated groups.

Lipid profile tests (Table 6 and Fig. 2D) revealed that the results displayed that *p* values for total cholesterol were (0.0004, 0.0066, and <0.0001) in (G2, G3, G4, and G5) correspondingly. In addition, HDL level was (<0.0001) in all groups of the treated animals. Moreover, *p* values for LDL level were (<0.0001, 0.0002, <0.0001) in (G3, G4, and G5), vs. to G2 showed a mildly insignificant difference. Furthermore, the triglyceride level was significantly increased in the treated groups compared to the control group *p*-values for (G2, G3, G4, and G5) were (0.0011, 0.0027, and <0.0001).

The liver function tests were performed (Table 6 and Fig. 2E). Parameters such as serum TSB, Albumin, protein, AP, ALP, and AST were measured. It was observed that the mean value of treated groups significantly decreased, by *p*-value (<0.0001). Whereas, Albumin levels in all treated groups were extremely increased *p* value (<0.0001). The mean value of protein level in experimental groups was significantly raised with a *p*-value (0.0133, 0.0055, 0.007, and 0.0005) respectively. The mean and standard deviation values of AP levels of treated groups (G2, G4, and G5) were significantly higher than the control, *p* values were (0.0011, 0.0271, and <0.0001) correspondingly. However, it was decreased in (G3) compared to G1.

*Attenuation of pathologic lesions by assessment of Lactuca serriola L. extract*

The liver section in the rats of control negative (G1) revealed normal arrangement and structures. Each hepatic lobule is centrally composed of a central vein that is surrounded by plate-like rows of hepatocytes that are separated by sinusoidal capillary with the portal area that shows normal histology without any pathologic alteration (Fig. 3a,b). Also, the hepatic sections showed intact histologic features in the G2 (Fig. 3c,d). While, mild vascular congestion and cellular swelling are characterized by wispy pale cytoplasm with a blurring lumen and centrally located nuclei in mild degree seen in the liver section of G3 (Fig. 3e,f). Microscopic features of the liver in G4 revealed mild-moderate pathologic alteration vs. to the previous groups and showed cloudy swelling of hepatocyte and vascular congestion particularly the central vein and sinusoidal capillary (Fig. 3g,h)., besides moderate congestion in G5, also hepatocytes showed centrilobular and perilobular microvesicular fatty degeneration of hepatocytes with focal-moderate inflammatory reaction specifically infiltration of neutrophils (Fig. 3i,j). Regarding score lesions, G5 showed the peak of it (score=10) followed by group G4 (score=3) vs. the groups G1 and G2 had the lowest one (score=0).

The microscopic features of the kidney in the control negative group (G1) showed intact histologic structures of glomeruli and collecting tubules with interstitial tubules without showing any alteration (Fig. 4a,b), vs. to the kidney section in groups G2 and G3 displayed minimum vascular congestion particularly in the glomerular tuft capillary and in G3 mild hemorrhage of the interstitial tissue detected, while normal features seen in the proximal and distal convoluted tubules (Fig. 4c-f). The renal sections of rats in G4 revealed (Fig. 4g,h), mild cellular swelling in the renal tubules with hemorrhage also in the renal interstitium and mild-moderate vascular congestion. While, the renal section in G5 has moderate cloudy swelling of tubules with blurred lumen, moderate vascular congestion, and interstitial hemorrhage (Fig. 4i,j). The renal score lesions achieved the greatest score in G5 (Score =6), followed by G4 with a score of 4, and the lowest score in G1 (score=0).

The microscopic sections of the spleens revealed intact splenic organization and histologic features of the parenchyma of white and red pulp in (G1, G2, and

G3) (Fig. 5a-f). In comparison, the splenic section in G4 showed mild follicular lymphocytic hyperplasia and white pulp expansion (Fig. 5g,h). While, the section of the spleen in G5 displayed moderate follicular lymphocytic hyperplasia with congestion in the red pulp (Fig. 5i,j). The splenic score lesions showed the lowest (scores=0) in the G1, G2, and G3 groups vs. the G5 group that recorded the highest score (scores=4).

## Discussion

Bioactive compounds, which are present in many different molecular configurations and may have therapeutic applications, are a distinctive and abundant source in herbs. Plants are used in traditional medicine to treat a wide range of disorders because they contain a significant number of physiologically active compounds that have therapeutic potential. Vegetables are nutritious because they include dietary fiber, vitamins, minerals, and phytochemicals. Adequate consumption of vegetables has been found to reduce the risk factors connected to certain chronic illnesses [13]. Our endeavor sought to ascertain the plant's lethal dose before moving forward with further research, based on earlier investigations owing to a dearth of data regarding *Lactuca serriola* L.

This study differs from others that have used *Lactuca serriola* at a dose of 6 g/kg in mice and did not cause any changes in body weight [14]. In the current study, the extract from *Lactuca serriola* affected body weight gain, and all the rats lost weight at the end of the experiment compared to the first day.

Table 3's evaluation of *Lactuca serriola's* effect on leucocyte levels revealed a decrease in leucocytes, including WBCs and lymphocytes, in the groups that received the higher dose of *Lactuca serriola*. This finding is consistent with prior studies that found Lactuca extract had anti-inflammatory properties because it contains triterpenoids and saponins [15, 16].

Although all treated groups had a substantial rise in MID% and GRA% as compared to the control group, the current results support previous research, particularly that Lactuca methanolic extract has an inhibitory inflammatory response [17, 18].

Comparing the Lactuca extract groups to the control group, the data relating to differential erythrocyte parameters (Table 4) showed non-significant alterations, which is not a significant difference. This finding is similar to the earlier finding that suggested this plant had cardiovascular protective properties [19].

With the exception of the mean platelet value being lower in the 800 mg/kg/bw Lactuca extracted group compared to the control group, all parameters were elevated in all treated groups, according to the differential thrombocyte counts disclosed in Table 5. Our rationale is the potent anticoagulant action of the Lactuca extract, whose clotting time of 110 s is similar to aspirin in the capillary tube method [20].

All blood lipid profiles, including total cholesterol, triglycerides, LDL, and HDL, were found to be significantly raised in the treated groups compared to the control group in this result (Table 6). This finding contradicts earlier research that found the plant to be protective against blood lipid improvement at doses of 200 and 500 mg/kg of *Lactuca serriola* aqueous extract [21, 22].

According to the results of the current study, which looked at the liver function biomarkers in Table 6, Lactuca extract significantly lowered the levels of TSB and ALT in all treated groups when compared to the control group. Another study found that the lactuca extract acts as a hepatoprotective and lowers the ALT activity, which is consistent with our research [23]. The level of protein, albumin, ALP, and AST in comparison to the control group increased in the Lactuca extract, contrary to our findings, other investigations demonstrated that the lactuca extract's antioxidant action resulted in a reduction of all liver enzyme indicators and protein [24-26].

The results of this study demonstrated that a higher dose of *Lactuca serriola* L caused toxicity and histologic alterations in the liver, kidney, and spleen. Rats treated with 400 and 800 mg/kg/bw showed moderately marked lesions, whereas the groups administered with 100 and 200 mg/kg/bw showed normal histologic structures, which are consistent with the previous result that documented the vasodilated effect of *Lactuca serriola*, which demonstrated the spasmogenic, spasmolytic, bronchodilator, and vasorelaxant properties of a 300 mg methanol extract from *Lactuca serriola* [19].

The current data disagrees with the findings of the study [27], research demonstrated that at 400 mg/kg, Lactuca leaf powder exhibited strong antiulcer properties. Previously, *Lactuca serriola* was used as a hepatoprotective drug at three distinct doses: 100, 250, and 500 mg/kg.bw, which is contrary to our findings. It is important to note that the plant was extracted for our investigation using methanol [28].

## Conclusion

The results demonstrated that oral administration of *Lactuca serriola* extracts at 100, 200, and 400 mg/kg bw for 14 days was safe for the rat model and did not result in any abnormal body weight measurements, behavioral changes, or histopathological findings; however, the 800 mg/kg bw dose was associated with noticeable negative effects.

*Conflicts of interest*

The authors declare no conflict of interest

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*Declaration of Conflict of Interest*

The authors declare that there is no conflict of interest.

*Ethical of approval*

The experimental structure was approved by the Ethical Committee of the College of Veterinary Medicine at the University of Sulaimani, which was done in compliance with the Guide for the Care and Use of Laboratory Animals (No: 030516).

**TABLE 1. Score assessment of the liver, kidney, and spleen's histological features.**

| Locations | Histopathologic abnormalities | Scores | Interpretation |
|---|---|---|---|
| **Blood vessels and Interstitial tissues** | **Congestion** | 0 | Absence of Change |
| | Mild | 1 | Change in less than 25% of parenchyma Change in 26-50% of parenchyma |
| | Moderate | 2 | |
| | Severe | 3 | Change in 51-100% of parenchyma |
| | **Hemorrhage** | 0 | Absence of Change |
| | Mild | 1 | Change in less than 25% of parenchyma Change in 26-50% of parenchyma |
| | Moderate | 2 | |
| | Severe | 3 | Change in 51-100% of parenchyma |
| **Cells** | **Swelling** | 0 | Absence of change |
| | Mild | 1 | Change in less than 25% of parenchyma Change in 26-50% of parenchyma |
| | Moderate | 2 | |
| | Severe | 3 | Change in 51-100% of parenchyma |
| | **Degeneration** | 4 | |
| **Parenchyma Inflammation** | **Inflammation** | 0 | No infiltration of cells |
| | Mild | 1 | Change in less than 25% of parenchyma Change in 26-50% of parenchyma |
| | Moderate | 2 | |
| | Severe | 3 | Change in 51-100% of parenchyma |
| **Parenchyma** | **Lymphocytic hyperplasia** | 0 | Absence of change |
| | Mild | 1 | Change in less than 25% of parenchyma Change in 26-50% of parenchyma |
| | Moderate | 2 | |
| | Severe | 3 | Change in 51-100% of parenchyma |

**TABLE 2. Body weight measurements (gram) in rats of all groups during the experiment.**

| Days | G1- Control (-Ve) | G2- 100mg/ml | G3- 200mg/ml | G4- 400mg/ml | G5- 800mg/ml |
|---|---|---|---|---|---|
| **0** | 150.67± 1.15 | 161.25± 1.50 | 183.50± 2.65 | 171.50± 5.74 | 184.50± 4.20* |
| **7** | 159.00± 3.61 | 161.00± 2.58 | 180.50± 6.86 | 172.25± 10.40 | 184.00± 4.69* |
| **14** | 157.33± 9.07 | 165.25± 2.87 | 173.00± 10.68 | 175.75± 13.23 | 164.50± 8.74* |

The value is expressed by Mean± STD. Within a column, values with star superscripts vary from each other ($P<0.05$).

**TABLE 3. The effect of *Lactuca serriola L.* extract on differential Leukocyte counts (gram) among studied groups.**

| *Parameters* | G1- Control (-Ve) | G2- 100mg/ml | G3- 200mg/ml | G4- 400mg/ml | G5- 800mg/ml |
|---|---|---|---|---|---|
| ***WBC ($10^9$/L)*** | 9.23± 0.64 | 14.35± 1.85* | 8.17± 2.18 | 5.70± 2.20 | 6.10± 1.00 |
| ***LYM ($10^9$/L)*** | 9.10± 0.17 | 8.95± 0.35 | 6.10± 1.21* | 3.75± 1.85* | 4.15± 0.35* |
| ***LYM (%)*** | 94.30± 1.80 | 62.25± 5.85* | 74.33± 7.09* | 61.75± 8.25* | 67.05± 4.85* |
| ***MID ($10^9$/L)*** | 0.63± 0.25 | 2.95± 0.65* | 1.57± 0.31* | 1.05± 0.25 | 1.00± 0.30 |
| ***MID (%)*** | 4.20± 0.36 | 20.90± 1.9* | 15.57± 1.90* | 20.00± 3.60* | 16.70± 2.01* |
| ***GRA ($10^9$/L)*** | 0.13± 0.06 | 2.45± 0.85* | 1.37± 0.40* | 0.90± 0.10 | 0.95± 0.35 |
| ***GRA (%)*** | 1.33± 0.25 | 16.85± 3.95* | 12.20± 0.82* | 18.20± 4.70* | 16.35± 2.85* |

The values are expressed by Mean± STD. Within a column, Leukocyte count values with star superscripts vary from each other ($P<0.05$). WBC=White blood cell; LYM=Lymphocyte; MID= Mid-range absolute counts; GRA= Granulocytes.

**TABLE 4. The effect of *Lactuca serriola L.* extracts on Erythrocyte counts (gram) among studied groups.**

| Parameters/ Unit | G1- Control (-Ve) | G2- 100mg/ml | G3- 200mg/ml | G4- 400mg/ml | G5- 800mg/ml |
|---|---|---|---|---|---|
| *RBC ($10^{12}$/L)* | 10.52± 0.58 | 9.82± 0.68 | 8.90± 1.22 | 8.64± 0.62 | 7.85± 0.87* |
| *HGB (g/dL)* | 19.43± 0.65 | 18.85± 1.45 | 16.03± 2.77 | 16.80± 1.40 | 14.90± 1.60* |
| *HCT (%)* | 56.00± 2.76 | 53.45± 3.75 | 42.07± 2.48* | 48.30± 4.50 | 43.85± 3.85* |
| *MCV (fL)* | 54.10± 0.00 | 54.40± 0.1 | 56.83± 5.32 | 55.80± 1.30 | 56.00± 1.20 |
| *MCH (pg)* | 18.63± 1.46 | 19.15± 0.15 | 20.80± 2.91 | 19.40± 0.30 | 19.05± 0.05 |
| *MCHC (g/Dl)* | 33.20± 1.23 | 35.20± 0.3 | 35.60± 1.75 | 34.80± 0.30 | 33.95± 0.65 |
| *RDWa (fl)* | 32.23± 1.96 | 31.20± 1.1 | 32.50± 4.41 | 32.20±0.40 | 32.80± 0.50 |
| *RDW (%)* | 13.60± 0.62 | 12.90± 0.6 | 13.03± 2.94 | 12.85± 0.35 | 13.20± 0.20 |

The values are expressed by Mean± STD. Within a column, each value with star superscripts varies from each other (P<0.05). RBC= Red blood cell; HGB= Hemoglobin; HCT= Hematocrit; MCV= Mean corpuscular volume; MCH= Mean corpuscular hemoglobin; MCHC= Mean corpuscular hemoglobin concentration; RDW= Red cell distribution width.

**TABLE 5. The effect of *Lactuca serriola L.* extract on differential thrombocyte counts among studied groups.**

| Parameters/ Unit | G1- Control (-Ve) | G2- 100mg/ml | G3- 200mg/ml | G4- 400mg/ml | G5- 800mg/ml |
|---|---|---|---|---|---|
| *PLT ($10^9$/L)* | 138.00± 5.57 | 553.50± 4.5* | 680.00± 3.61* | 672.33± 6.51* | 400.33± 6.03* |
| *MPV (fL)* | 7.43± 0.76 | 7.15± 0.45 | 7.67± 0.45 | 7.05± 0.05 | 6.34± 0.29* |
| *PDWa (fL)* | 8.43± 1.42 | 8.85± 0.75 | 8.93± 0.25 | 8.43± 0.12 | 8.14± 0.24 |
| *PDW (%)* | 35.53± 4.65 | 36.95± 2.75 | 36.10± 1.35 | 35.55± 0.45 | 33.93± 1.42 |
| *PCT (%)* | 0.09± 0.02 | 0.21± 0.165 | 0.43± 0.06* | 0.34± 0.19 | 0.53± 0.04* |
| *P- LCR (%)* | 8.53± 0.67 | 10.55± 2.978 | 10.27± 0.75 | 8.10± 0.90 | 5.57± 0.51 |
| *P-LCC ($10^9$/L)* | 11.00± 2.00 | 22.52± 3.75* | 63.93± 8.10* | 50.27± 3.37* | 42.90± 1.35* |

The values are expressed by Mean± STD. Within a column, each value with star superscripts varies from each other (P<0.05). PLT=Platelet; MPV=Mean platelet volume; PCT=Platelet hematocrit; P- LCR= Platelet-large cell ratio; P-LCC= Platelet-large cell ratio.

**TABLE 6. The effect of *Lactuca serriola L.* extract on blood lipid profile and liver biomarkers among studied groups.**

| *Parameters/ Unit* | G1- Control (-Ve) | G2- 100mg/ml | G3- 200mg/ml | G4- 400mg/ml | G5- 800mg/ml |
|---|---|---|---|---|---|
| *Total Cholesterol (mg/dL)* | 19.13± 1.51 | 33.10± 5.08* | 28.62± 1.74* | 37.11± 2.06* | 81.13± 1.99* |
| *TG (mg/dL)* | 32.12± 2.44 | 47.61± 6.52* | 45.81± 1.50* | 58.13± 2.01* | 57.79± 2.80* |
| *HDL (mg/dL)* | 5.31± 0.79 | 21.30± 3.58* | 22.63± 2.53* | 25.75± 1.57* | 62.58± 2.49* |
| *LDL (mg/dL)* | 1.82± 0.95 | 4.44± 1.10 | 9.97± 1.30* | 8.09± 1.06* | 13.77± 1.32* |
| *Total Serum Bilirubin(mg/dL)* | 0.13± 0.03 | 0.10± 0.00 | 0.10± 0.00 | 0.00± 0.00* | 0.00± 0.00* |
| *Albumin (g/dL)* | 1.63± 0.15 | 3.55± 0.15* | 3.80± 0.40* | 3.53± 0.35* | 4.37± 0.31* |
| *Protein (g/dL)* | 3.70± 0.92 | 6.10± 0.40* | 6.47± 1.26* | 6.37± 0.67* | 7.53± 0.35* |
| *ALP (U/L)* | 14.70± 1.25 | 71.55± 27.95* | 49.13± 4.99* | 144.00± 4.01* | 200.03± 3.20* |
| *ALT (U/L)* | 178.20± 4.45 | 57.60± 2.50* | 40.83± 2.58* | 67.00± 2.00* | 83.47± 3.90* |
| *AST (U/L)* | 310.50± 4.85 | 327.00± 4.39* | 211.07± 2.70* | 398.63± 2.60* | 691.80± 2.79* |

The values are expressed by Mean± STD. Within a column, each value with star superscripts varies from each other (P<0.05). TG= Triglycerides, HDL= High-density lipoprotein, LDL= Low-density lipoprotein, ALP= Alkaline phosphatase, AST= Aspartate aminotransferase, ALT= Alanine aminotransferase.

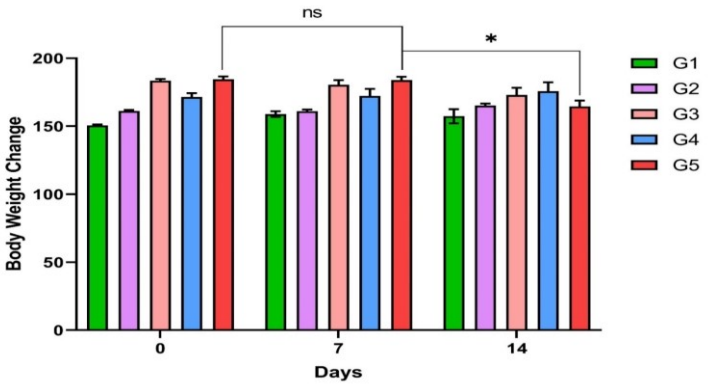


**Fig. 1. The body weight changes in studied groups.**

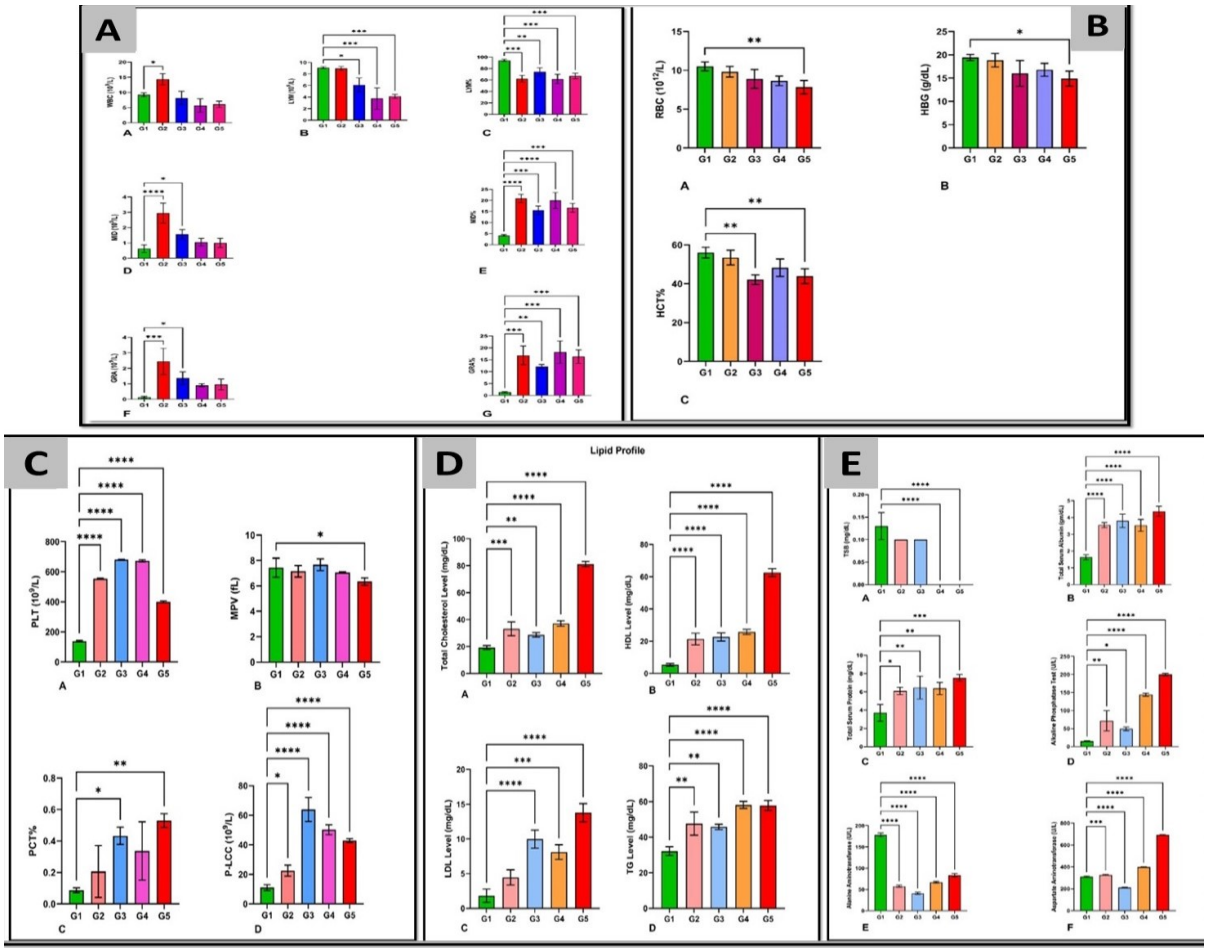


**Fig. 2. The effect of the Lactuca serriola L. extract on variable blood parameters count in control and treated groups; A: Differential leukocyte counts, B: Differential erythrocyte counts, C: Differential thrombocyte counts, D: Lipid profile levels, E: Liver biomarkers level.**

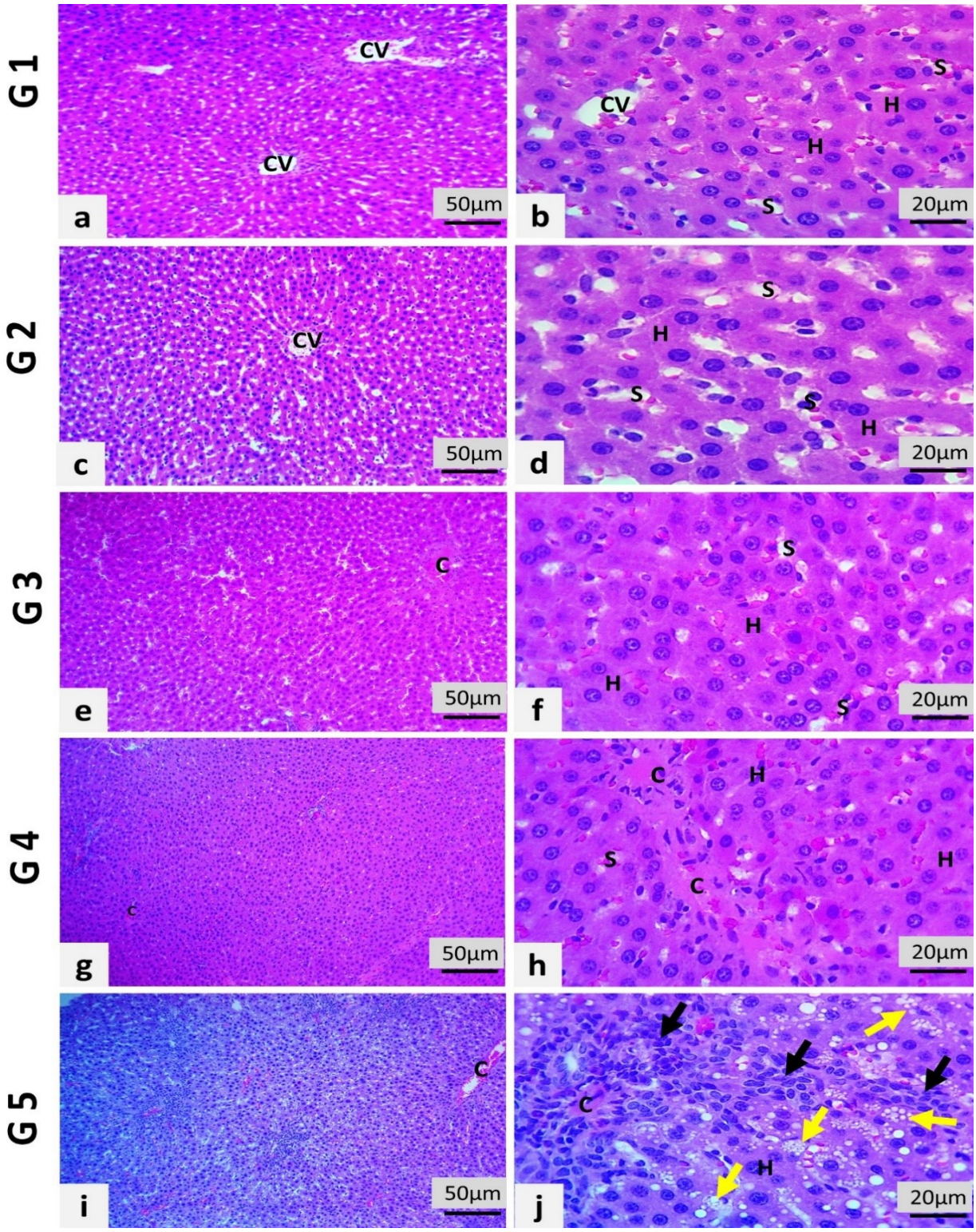


**Fig. 3. Microscopic section of the liver in rat treated with different doses of *Lactuca serriola L.;* a and b: Normal histologic appearance of the liver that showed central vein (CV) surrounded by normal hepatocytes (H) plate and sinusoidal capillary (S) that separate the hepatocytes in G1. C and d: Normal histologic structures of liver parenchyma in G2. E and f: Mild swelling of hepatocytes with mild congestion of central vein I in G3. G and h: Moderate congestion I of central vein and sinusoidal capillary with mild hepatocyte swelling in G4. I and j: Moderate congestion I of central vein, microvesicular fatty degeneration of hepatocyte (yellow arrows), and focal inflammatory reaction (black arrows) in G5, (H & E stain).**

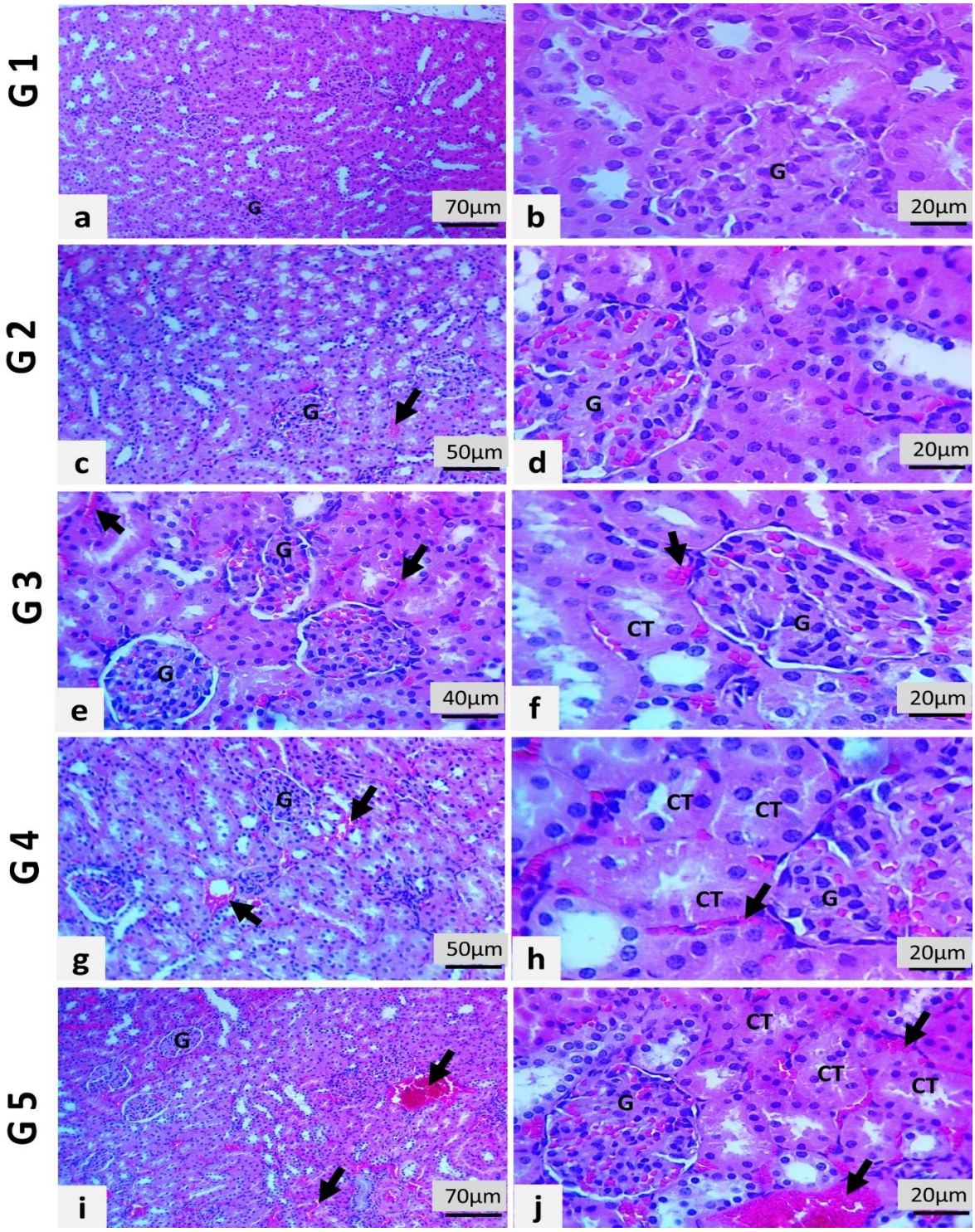


**Fig. 4. Microscopic section of kidney in rat treated with different doses of *Lactuca serriola L.;* a and b: Normal histologic structures of glomeruli (G) with intact collecting tubules (CT) in G1. C and d: Mild congestion of glomerular capillary and renal vasculature (black arrows), with normal morphology of CT in G2. E and f: Mild congestion of glomeruli with mild hemorrhage of interstitial tubules (black arrows) in G3. G and h: Mild-moderate congestion of glomeruli with hemorrhage of interstitial tubules (black arrows), and mild swelling of CT in G4. I and j: Moderate congestion of nephron and renal vasculature with hemorrhage of interstitial tubules (black arrows), and moderate swelling of CT in G5, (H & E stain).**

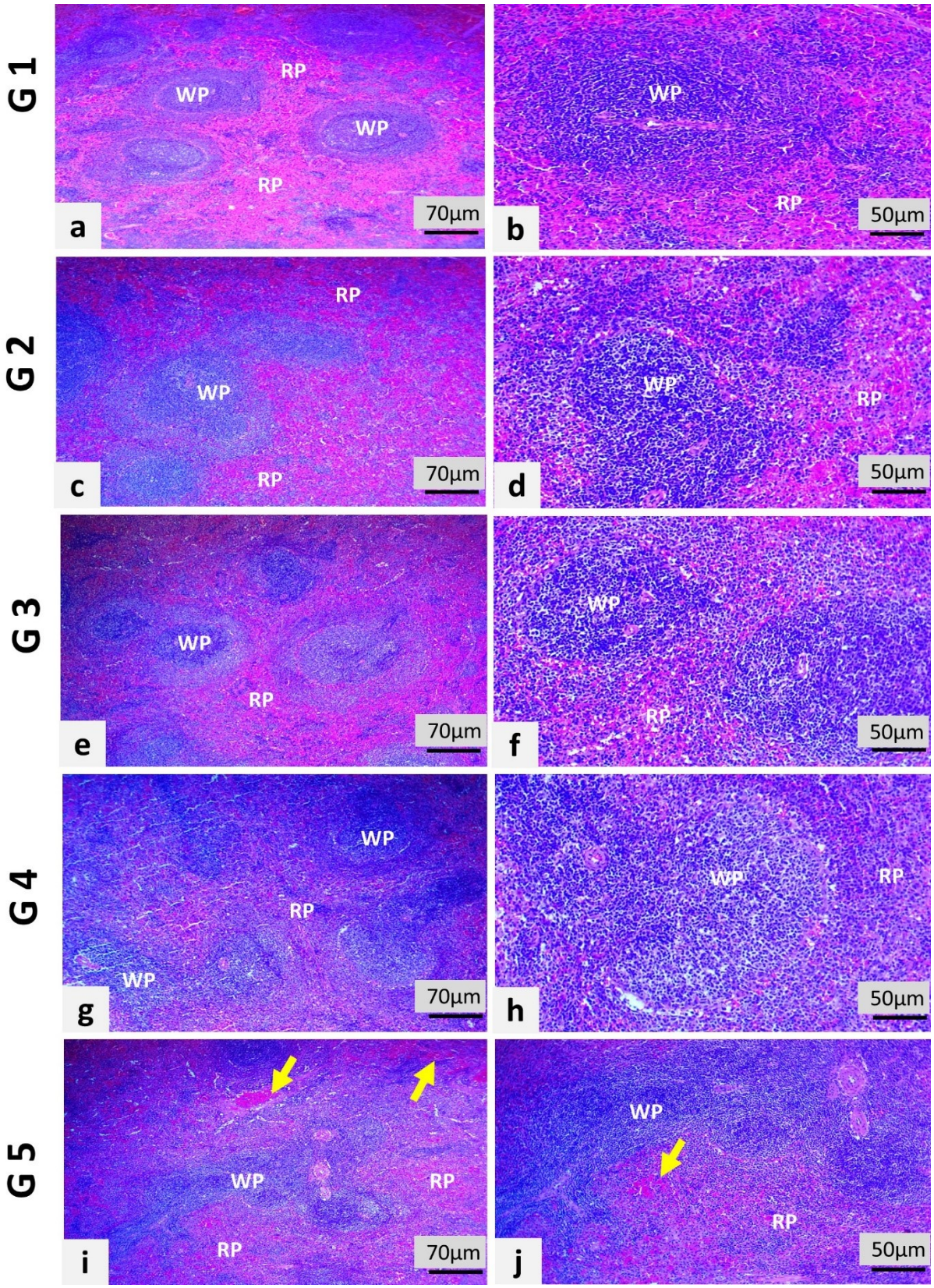


**Fig. 5. Microscopic section of the spleen in rat treated with different doses of *Lactuca serriola L.;* a and b: Normal splenic parenchyma, white pulp (WP), and red pulp (RP) in G1. C-f: Intact splenic organization in G2 and G3 respectively. g and h: Mild follicular lymphocytic hyperplasia in G4. I and j: Moderate follicular lymphocytic hyperplasia with congestion in red pulp (yellow arrows) in G5, (H & E stain).**

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## تقييم التأثير السمي لنبات *Lactuca serriola L.* (خَسّ الزَّيْت) في الجرزان

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**الملخص**

تهدف هذه الدراسة الى تقييم المستويات السمية للمستخلص الكحولي لنبات خَسّ الزَّيْت، شملت الدراسة الى استخدام ثلاثون من الجرذان البالغة قسمت الى خمس مجموعات رئيسية أولى هذه المجموعات مجموعة السيطرة او المجموعة السالبة أي التي لم تتلقى أي نوع من العلاجات، أما المجاميع من الثانية الى الخامسة فهي مجاميع العلاج التي اعطي لها باستخدام أنبوب التجريع ، وكانت جرعات التجريع على النحو التالي (400,200,100) ملغم /كغم من الوزن الحي على التوالي مدة الدراسة كانت أربعة عشر يوميا أخذت فيها عينات الدم لغرض اجراء الفحوصات البيوكيميائية وكما أخذت مسحات نسيجية من الكلى والكبد والطحال بعد صبغها بصبغة الايوزين والهيماتوكسلين ، أظهرت الدراسة عدم وجود تفاوت أو فرق في اوزان الجسم لجميع المجموعات بينما لوحظ انخفاض واضح في عدد الكريات البيضاء وخلايا اللمفاوية وبالأخص في المجموعة التي استلمت جرعات عالية من نبات خَسّ الزَّيْت وبالإضافة أدى استخدام خَسّ الزَّيْت إلى زيادة كبيرة في عدد الصفائح الدموية التفاضلية، وجميع مؤشرات الدهون في الدم، وعلامات إنزيمات الكبد مقارنة بمجموعة السيطرة

كما أظهرت الفئران التي تلقت جرعات 400 و800 ملغم /كجم/وزن الجسم تلفًا نسيجيًا كبيرًا في الكبد والكلى والطحال، وعلى النقيض من ذلك، كان لدى الفئران التي أعطيت جرعات 100 و200 مجم/كجم/وزن الجسم كانت جميع الأعضاء المذكورة أنفأ بحالة طبيعية، هذه النتائج وفرت معلومات قيمة حول الجرع الأمنه والتي تتراوح ما بين (100-400) ملغم للمركبات الفينولية المستخرجة من نبات خَسّ الزَّيْت لأجراء تجارب بحثية في الحيوانات.

**الكلمات المفتاحية**: التنكس الدهني ، خَسّ الزَّيْت ، إنزيم الكبد ، تضخم اللمفاويات ، منطقة شاربازهر.