



## Impact of *Eruca sativa* Leaves Aqueous Extract on Liver Function, Immunity Profile and Behavioural Responses of Healthy and Thioacetamide Intoxicated Male Albino Mice

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### ABSTRACT

The current experiment was intended to investigate the hepatoprotective, immunostimulant activity, behavioural and anxiety responses of mice supplemented with *Eruca sativa* aqueous extract (ESAE) at a dose of 1% in drinking water in healthy and thioacetamide (TAA) intoxicated male mice. Eighty male albino mice were divided into four treatments of 20 mice each, with two replicates. The T1 considered as control group, T2 was the TAA intoxicated group, T3 was supplemented with ESAE only in drinking water for 3 weeks, and T4 supplemented with both ESAE for 3 weeks and TAA in the last 3 days in drinking water. The effect of ESAE was assessed by estimation of erythrogram, leukogram, platelet count, N/L ratio, total protein, albumin, globulin, and A/G ratio, and by evaluating the activity of liver enzymes such as aspartate transaminase (AST) and alanine transaminase (ALT) as well as the behavioural responses and anxiety of mice were also recorded. ESAE administration markedly prevents the elevation of plasma liver enzymes after oral administration of TAA. The mice supplemented with ESAE (T3) had higher mean values of total protein, globulin, with lower neutrophils count and A/G ratio compared to the control group (T1) and TAA intoxicated mice (T2), indicating that ESAE has immunostimulant and hepatoprotective activity. The mice supplemented with ESAE (T3) groomed, scratched, and sniffed significantly more than mice of T2, with significantly less resting and sleeping. Regarding the elevated plus maze results; when comparing mice of T1 and T2 with other treatments, the mice of T3 and T4 showed a huge ( $P < 0.05$ ) decline in the quantity of closed arms entries, with a relating increase in the quantity of open arms entries. It is concluded that a daily oral dose of 1% ESAE improves mouse activity, reduces anxiety, and has immunostimulant and hepatoprotective activity.

### Original Article:

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The liver is considered the vital organ that regulates the metabolic and physiological homeostasis of the body (Verma and Khosa, 2010). The liver's essential capacities include sugar, protein, fat digestion, detoxification; bile secretion; and nutrient stockpiling. It partakes in all biochemical development pathways, illness opposition, and supplement supply. Thus, keeping a sound liver status is a significant component for generally speaking, well-being and welfare (Rajib *et al.*, 2009).

In recent years, great attention has been given to herbal medicine due to its low cost and greater compatibility (Azadbakht *et al.*, 2003). However, the synthetic agents have severe undesirable side effects. The hepatoprotective property of the body have been improved through using of various plants that improve the antioxidant status (Vinothkumar *et al.*, 2010). In this way, there is a lot of consideration regarding assessing the logical premise of conventional herbal drugs that have hepatoprotective action (Shahani, 1999).

Many home-grown plants, including *Eruca sativa* (Family: Cruciferae), have been read up for use in different liver issues, which control oxidative pressure because of their cancer prevention agent properties. Fresh *Eruca sativa* has a trademark solid flavor that is believed to be connected with the presence of glucosinolates and their breakdown items as isothiocyanates (Bennett et al., 2006), which have numerous organic exercises, including anticarcinogenic, antifungal, antibacterial, and cancer prevention agent impacts (Kim et al., 2004). Alam et al., (2007) announced that the leaves and seeds of *Eruca sativa* have powerful radical scavenging antioxidant action and increment or keep up the levels of antioxidant agents and antioxidant enzymes to safeguard the body against oxidative harm.

Thioacetamide (TAA) is a powerful hepatotoxin that is utilized to control the crumbling of organic citrus products, particularly oranges, so it is utilized as a fungicide. TAA is a sulfur donor compound and can be used in many industrial and drug applications. Additionally, TAA is a cancer-causing compound (Chilakapati et al., 2005). TAA can initiate neurotic cirrhosis of liver, which is similar to human liver cirrhosis caused by alcohol in many elements. TAA is one of the various factors that causes structural and functional changes in liver and different tissues like the kidney, thymus, spleen, and lungs (Sirag, 2007). On delayed openness, TAA prompts the development of hyperplastic knobs, cell adenomas, hepatocarcinoma, and cirrhosis. TAA-instigated cirrhosis in rodents has been demonstrated to be a reasonable test model of this sickness with an etiology and pathology equivalent to those found in people (Yeh et al., 2004).

The elevated plus maze (EPM) has been depicted as a straightforward strategy for evaluating the anxiety reactions of rodents. Furthermore, the anxiolytic and anxiogenic impacts of pharmacological specialists, drug abuse, and chemicals can be explored by the elevated plus-maze test (Walf and Frye, 2007). The appraisal of anxiety reactions of rodents relies upon the time spent on the open arms and the time spent on the closed arms, which depends upon the introduction of harmful stimuli. The quantity of closed arm entries is viewed as a guide of general motor activity. In any case, the quantity of open-arm entries is connected with both anxiety and general activity and is a blended guide (Darwish et al., 2001).

Because of the absence of defensive medications in treating liver infections, medicinal plants have become profoundly famous (Mohammed et al., 2009). Accordingly, the point of this study was to assess the prophylactic impact of *Eruca sativa* leaves extract (ESAE) in healthy and thioacetamide intoxicated (TAA) male mice.

## MATERIALS AND METHODS

All events of animal handling and samples' assembly and discarding were as per the Institutional Animal Care and Use Committee (IACUC) guidelines with oversight of the office of Veterinary Medicine, University of Sadat City (VUSC-020-1-20).

### 1. Chemicals:

Thioacetamide (TAA) was bought from Sigma-Aldrich (St. Louis, MO). All examination kits were purchased from Al Gomhoria Company, Cairo, Egypt. Any remaining chemicals were of scientific grade. Thioacetamide was prepared freshly by dissolving precious stones in sterile refined water and blending until all stones were broken up.

### 2. Plant material and extraction:

The leaves of *Eruca sativa* leaves were purchased from the public market of Sadat City, Menofia Governorate, Egypt. Fluid concentrate of *Eruca sativa* leaves was ready as per the strategy portrayed by Grami et al., (2019). The concentrate was sifted and put away in a dim, sterile jug at 4°C until utilized.

### 3. Animal housing and accommodation:

Eighty male albino mice weighing 28-39 g were bought from the Husbandry and Animal wealth department, Faculty of Veterinary Medicine, University of Sadat City, Menofia, Egypt. They were housed in polypropylene cages floored with sawdust under stable circumstances like temperature (21°C), humidity (40%), and a standard 12-hour light and 12-hour dark cycle. They were given a pelleted diet and water *ad libitum*.

### 4. Experimental design:

After one week of adaptation, the mice were partitioned into four treatments of 20 mice each, with two replicates as displayed in Table (1).

Table 1: Experimental design:

Treatments	Water	TAA	ESAE
T1	<i>Ad libitum</i>	-	-
T2	-	300 mg/L	-
T3	-	-	1 %
T4	-	300 mg/L	1 %

The ESAE were supplemented to mice in drinking water for 21 days; TAA was also administered to mice in drinking water at the last three days of ESAE supplementation (at 19<sup>th</sup>, 20<sup>th</sup>, 21<sup>st</sup> day).

#### 4.1. Mice behavioural responses:

In each cage of the four treatments, the mice were observed for 10 minutes in two observational periods: in the morning (9:00-10:20) and the afternoon (14:00-15:20). The behaviour of mice in each

treatment was recorded using a video camera. Data was collected from the video recordings using instantaneous scan sampling (Martin and Bateson, 1993). The percentage of mice that performed resting and sleeping (inactivity), grooming and scratching (activity), and sniffing and rearing (exploration) behaviour was recorded during all scan samples in each treatment.

**4.2. The elevated plus maze test (Anxiety test):**

In the 22<sup>nd</sup> day (after TAA administration), the elevated plus maze (EPM) examination is utilized to gauge four mice's anxiety per group. The wooden contraption comprises two restricting arms: the open arms (5 × 35 cm) and the closed arms (5 × 35 × 30 cm). The mouse was set toward the finish of an open arm, confronting the central platform, and permitted to investigate the apparatus for 5 minutes and observed using a video camera (Sony - Japan) (Komada, 2008). After every trial, the maze was cleaned with a material dunked in 70% ethyl liquor and permitted to dry. The test was performed somewhere in the range of 13:00 and 14:20 hrs. The complete time spent in the closed and open arms (sec) and the entries numbers into the shut and open arms were recorded.

**4.3. Hematological parameters (Erythrogram, leukogram):**

In the 22<sup>nd</sup> day (after TAA administration), the mice abstained for 12 h, gauged and exposed to light chloroform sedation, ten blood samples were gathered via cardiac puncture on heparinized tubes for assurance of complete blood count (CBC) utilizing an Automatic blood analyzer (Abbott Diagnostic Division, Canada) as the accompanying boundaries: leucocytes (WBCs),

erythrocytes (RBCs), lymphocytes count, neutrophils count, N/L ratio, hemoglobin (Hb), hematocrit volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and platelets count.

**4.4. Biochemical parameters:**

The samples of blood were centrifuged at 3000 rpm for 10 min to isolate plasma and kept at -20°C until used to determine the following parameters.

**4.4.1. Total protein, albumin, and globulin:**

Total protein and albumin are set in stone by utilizing commercial kits bought from Diamond Diagnostics (Doumas, 1975; Doumas, et al., 1971). Globulin levels were determined by subtracting albumin values from total protein.

**4.4.2. Aspartate aminotransferase (AST) and alanine aminotransferase (ALT):**

Entirely settled by utilizing commercial kits bought from BioMed Diagnostics (Reitman and Frankel, 1957).

**4.5. Statistical analysis:**

The data accumulated during the examination was communicated as the mean ± SE of the absolute number of animals utilized in each group. Information was exposed to one-way analysis of variance (ANOVA) and Duncan multiple range tests to decide the distinction between the exploratory and control groups. A degree of the meaning of P < 0.05 was viewed as measurably significant.

**RESULTS**

**1. Mice behavioural responses:**

In Table (2), the effects of ESAE and TAA on the behavioural responses of mice are presented. The mice of T2 supplemented with TAA only presented a critical (P<0.05) high inactivity percentage as resting & sleeping compared to those of T3 supplemented with ESAE only. On the other hand, TAA supplementation (T2) significantly reduced the percentage of mice activity as grooming and scratching behaviour compared to mice of T3 supplemented ESAE only and T4 supplemented ESAE followed by TAA. Mice from the ESAE group (T3) sniffed significantly more than mice from the other groups.

Table 2: The impact of aqueous extract of *Eruca sativa* leaves on the behavioural responses (%) of healthy and thioacetamide intoxicated mice (mean ± SE).

Behavioral patterns (%)	Treatments			
	T1	T2	T3	T4
<b>Inactivity</b>				
Resting	0.11±0.05 <sup>ab</sup>	0.24±0.04 <sup>a</sup>	0.08±0.02 <sup>b</sup>	0.12±0.05 <sup>ab</sup>
Sleeping	0.23±0.06 <sup>ab</sup>	0.31±0.05 <sup>a</sup>	0.13±0.03 <sup>b</sup>	0.21±0.05 <sup>ab</sup>
<b>Activity</b>				
Grooming	0.17±0.04 <sup>ab</sup>	0.08±0.02 <sup>b</sup>	0.29±0.04 <sup>a</sup>	0.26±0.04 <sup>a</sup>
Scratching	0.09±0.03 <sup>ab</sup>	0.007±0.007 <sup>b</sup>	0.14±0.03 <sup>a</sup>	0.12±0.03 <sup>a</sup>
<b>Exploration</b>				
Sniffing	0.07±0.02 <sup>b</sup>	0.10±0.02 <sup>b</sup>	0.23±0.03 <sup>a</sup>	0.10±0.03 <sup>b</sup>
Rearing	0.06±0.02	0.06±0.01	0.06±0.01	0.08±0.02

In the same raw, <sup>a, b, c</sup> Means ± SE with various letters superscripts are essentially different at (P < 0.05).

T1: control    T2: TAA only    T3: ESAE only    T4: ESAE+TAA.

## 2. Anxiety test:

The impacts of ESAE on the anxiety of mice are introduced in Table (3). Mice treated with ESAE showed a significant ( $P < 0.05$ ) decrease in stress and anxiety. While looking at the mice of T1 and T2 with other treatments, it is observed that the mice of T3 and T4 had a significant ( $P < 0.05$ ) decline in the quantity of closed arms entries and an increase in the quantity of open arms entries. Moreover, mice of T2 invested essentially more times in closed arms than mice of T1, T3, and T4. T3 mice, on the other hand, invested fundamentally additional time in the open arm than T2 mice.

Table 3: The impact of aqueous extract of *Eruca sativa* leaves on healthy and thioacetamide intoxicated mice anxiety (mean  $\pm$  SE):

Items	Treatments			
	T1	T2	T3	T4
Numbers of entries into closed arms	3.00 $\pm$ 0.57 <sup>a</sup>	3.33 $\pm$ 0.33 <sup>a</sup>	1.33 $\pm$ 0.33 <sup>b</sup>	1.33 $\pm$ 0.33 <sup>b</sup>
Time spent in the closed arm (sec)	105.66 $\pm$ 1.60 <sup>b</sup>	146.66 $\pm$ 1.02 <sup>a</sup>	29.33 $\pm$ 3.66 <sup>c</sup>	40.33 $\pm$ 1.45 <sup>c</sup>
Numbers of entries in the open arms	1.66 $\pm$ 0.33 <sup>b</sup>	1.66 $\pm$ 0.33 <sup>b</sup>	2.33 $\pm$ 0.33 <sup>a</sup>	2.00 $\pm$ 0.14 <sup>a</sup>
Time spent in the open arm (sec)	54.66 $\pm$ 1.08 <sup>ab</sup>	30.00 $\pm$ 1.02 <sup>b</sup>	60.00 $\pm$ 1.54 <sup>a</sup>	57.00 $\pm$ 1.73 <sup>ab</sup>

In the same raw, <sup>a, b, c</sup> Means  $\pm$  SE with various letters superscripts are essentially different at ( $P < 0.05$ ).

T1: control T2: TAA only T3: ESAE only T4: ESAE+TAA.

## 3. Hematological parameters:

Table (4) revealed non-significant ( $P > 0.05$ ) effects on the erythrocyte profile of all treatments when compared with control group T1 indicating the safety of ESAE administration. The oral administration of TAA for mice of T2 showed a significant ( $P < 0.05$ ) increase in WBC count, neutrophil count, and N/L ratio. Both the administration of ESAE only in T3 and ESAE followed by TAA in T4 showed a significant ( $P < 0.05$ ) increase in neutrophil count, N/L ratio, and platelet count as displayed in Table (4).

Table 4: The impact of aqueous extract of *Eruca sativa* leaves on hematological parameters of healthy and thioacetamide intoxicated mice (Mean  $\pm$  SE).

Items	Treatments			
	T1	T2	T3	T4
<b>Erythrocyte parameters</b>				
RBCs $\times 10^6/\text{mm}^3$	5.21 $\pm$ 0.50 <sup>a</sup>	5.73 $\pm$ 0.25 <sup>a</sup>	5.53 $\pm$ 0.40 <sup>a</sup>	5.68 $\pm$ 0.40 <sup>a</sup>
Hb g/dl	10.40 $\pm$ 0.85 <sup>a</sup>	11.00 $\pm$ 0.44 <sup>a</sup>	10.73 $\pm$ 0.89 <sup>a</sup>	10.63 $\pm$ 0.71 <sup>a</sup>
PCV %	31.73 $\pm$ 1.77 <sup>a</sup>	32.33 $\pm$ 0.85 <sup>a</sup>	31.67 $\pm$ 1.77 <sup>a</sup>	32.01 $\pm$ 1.61 <sup>a</sup>
MCHC %	32.78 $\pm$ 0.80 <sup>a</sup>	34.02 $\pm$ 0.74 <sup>a</sup>	33.88 $\pm$ 1.09 <sup>a</sup>	33.20 $\pm$ 0.43 <sup>a</sup>
MCH (pg)	19.96 $\pm$ 0.22 <sup>a</sup>	19.20 $\pm$ 0.42 <sup>a</sup>	19.40 $\pm$ 0.35 <sup>a</sup>	18.72 $\pm$ 0.06 <sup>a</sup>
MCV (fl)	60.90 $\pm$ 0.77 <sup>a</sup>	56.42 $\pm$ 0.28 <sup>a</sup>	57.27 $\pm$ 0.35 <sup>a</sup>	56.36 $\pm$ 0.24 <sup>a</sup>
<b>Leucocyte's parameters</b>				
WBCs ( $10^3/\text{mm}^3$ )	9.27 $\pm$ 0.37 <sup>b</sup>	14.27 $\pm$ 2.69 <sup>a</sup>	10.03 $\pm$ 0.36 <sup>ab</sup>	10.57 $\pm$ 1.23 <sup>ab</sup>
Lymphocytes $10^3/\text{mm}^3$	5.97 $\pm$ 0.11 <sup>a</sup>	6.00 $\pm$ 0.35 <sup>a</sup>	5.77 $\pm$ 0.56 <sup>a</sup>	5.71 $\pm$ 0.87 <sup>a</sup>
Neutrophils $10^3/\text{mm}^3$	2.50 $\pm$ 0.26 <sup>c</sup>	5.47 $\pm$ 1.09 <sup>a</sup>	3.17 $\pm$ 0.23 <sup>b</sup>	3.07 $\pm$ 2.11 <sup>b</sup>
N/L ratio	0.42 $\pm$ 0.05 <sup>c</sup>	0.91 $\pm$ 0.08 <sup>a</sup>	0.55 $\pm$ 0.03 <sup>b</sup>	0.54 $\pm$ 0.05 <sup>b</sup>
Platelets count $10^3/\text{mm}^3$	543.00 $\pm$ 24.14 <sup>b</sup>	486.00 $\pm$ 33.92 <sup>c</sup>	697.67 $\pm$ 27.52 <sup>a</sup>	711.67 $\pm$ 29.98 <sup>a</sup>

In the same raw, <sup>a, b, c</sup> Means  $\pm$  SE with various letters superscripts are essentially different at ( $P < 0.05$ ).

T1: control T2: TAA only T3: ESAE only T4: ESAE+TAA.

## 4. Biochemical parameters:

Table (5) showed a significant ( $P < 0.05$ ) increase in total protein and globulin in T3 and T4 contrasted with T1 and T2. Nonetheless, there was a critical diminishing in the A/G ratio in T3 contrasted with T2. The effects of oral supplementation of ESAE for 21 days and TAA on liver enzymes in male mice are addressed in Table (5). The outcomes uncovered a significant ( $P < 0.05$ ) increase in plasma ALT and AST action in T2 and T4

contrasted with T1 and T3. While mice of T3 showed a non-significant effect ( $P>0.05$ ) in AST action when contrasted with mice of T1, demonstrating the hepatoprotective impact of ESAE.

Table 5: The impact of aqueous extract of *Eruca sativa* leaves on biochemical parameters of healthy and thioacetamide intoxicated mice (Mean  $\pm$  SE).

Items	Treatments			
	T1	T2	T3	T4
<b>Total protein (g/dl)</b>	3.94 $\pm$ 0.12 <sup>b</sup>	3.98 $\pm$ 0.25 <sup>b</sup>	5.20 $\pm$ 0.36 <sup>a</sup>	4.71 $\pm$ 0.29 <sup>a</sup>
<b>Albumin (g/dl)</b>	2.30 $\pm$ 0.09 <sup>a</sup>	2.42 $\pm$ 0.10 <sup>a</sup>	2.42 $\pm$ 0.18 <sup>a</sup>	2.55 $\pm$ 0.13 <sup>a</sup>
<b>Globulin (g/dl)</b>	1.64 $\pm$ 0.09 <sup>c</sup>	1.56 $\pm$ 0.16 <sup>c</sup>	2.78 $\pm$ 0.45 <sup>a</sup>	2.16 $\pm$ 0.22 <sup>b</sup>
<b>A/G ratio</b>	1.44 $\pm$ 0.10 <sup>ab</sup>	1.65 $\pm$ 0.13 <sup>a</sup>	1.08 $\pm$ 0.24 <sup>b</sup>	1.29 $\pm$ 0.16 <sup>ab</sup>
<b>ALT (U/L)</b>	30.17 $\pm$ 3.40 <sup>c</sup>	259.33 $\pm$ 29.58 <sup>a</sup>	48.00 $\pm$ 1.76 <sup>c</sup>	105.20 $\pm$ 9.59 <sup>b</sup>
<b>AST (U/L)</b>	94.43 $\pm$ 4.01 <sup>c</sup>	307.67 $\pm$ 14.87 <sup>a</sup>	116.00 $\pm$ 3.61 <sup>bc</sup>	121.17 $\pm$ 8.87 <sup>b</sup>

In the same raw, <sup>a, b, c</sup> = Means  $\pm$  SE with various letters superscripts are essentially different at ( $P < 0.05$ ).  
 T1: control    T2: TAA only    T3: ESAE only    T4: ESAE+TAA.

### DISCUSSION

In the past decade, herbal plants and alternative medicines have been used to treat various disorders as their toxicity features seem to have inferior side effects (Majid, et al., 2011). The liver assumes a positive part in most metabolic cycles, explicitly detoxification. The liver kills a wide scope of poisonous synthetic compounds, both those created inside and those approaching from the outside environment (Chiang, 2014). Hepatotoxic agents cause structural abnormalities in the liver, altering normal physiological functions (ELSadek, 2014). So, when the liver is healthy, it reflects on the animal's health, activity, and welfare.

No literature studies the effects of ESAE on animal behaviour and the anxiety of intoxicated mice by TAA. The results of the behavioural responses of mice indicated that TAA intoxicated mice (T2) showed a high percentage of inactivity as resting and sleeping, with a low percentage of grooming and scratching in comparison to those of mice of T3 supplemented with ESAE only. Likewise, results from an anxiety test (EPM) demonstrated that TAA organization incites an anxiogenic or upsetting impact by expanding the number of passages in the closed arm with a diminishing number of sections in the open arms. As well as the stressful effect of TAA, indicated by the higher N/L ratio.

These results may be attributed to TAA supplementation's ability to increase the rate of brain and liver oxidative stress, which leads to hepatic failure and neurotoxicity. These results supported Tunes et al., (2005) indication that a single dose

of thioacetamide (150mg/kg) in Wistar rats induces hepatic failure and brain oxidative stress. Therefore, TAA supplementation decreased mouse activity and increased the stress effect on mice, indicated by less grooming mainly. This result agreed with Moyaho and Valencia (2002), who reasoned that preparing has for some time been viewed as a social marker of stress and Choleris et al., (2001) detailed that prepping is exceptionally touchy to different stressors. As well as Cromwell et al., (1998), Kalueff and Tuohimaa, (2005) clarified that numerous districts in mind have all the earmarks of being associated with the guideline of preparing practices.

It is an exciting finding that ESAE may eliminate the toxic effect of TAA. The unfortunate impact was diminished by ESAE supplementation for TAA inebriated mice, as determined by critical reductions in the quantity of closed arms entries, with a comparing expansion in the quantity of open arms entries of the EPM test. Mechiel Korte and De Boer, (2003) announced absolute sections and time spent in each arm of EPM, establish the record of essential tension. In this way, evasion of open arms is viewed because of the acceptance of more elevated levels of dread (Rodgers and Dalvi, 1997). It is believed that the aversion of mice to investigating the open arms of the labyrinth is brought about by their anxiety toward open and raised spaces (Komada., 2008).

Ethanollic & aqueous extracts of *Eruca Sativa* demonstrate hepatoprotective effects against liver toxicity (Mashi et al., 2017). Moreover, our results revealed that ESAE supplementation for mice had a hepatoprotective effect against the toxic effects of TAA. Likewise, ESAE improved the mice's immunity,

which enabled them to withstand the stressful impact of TAA, as indicated by the decreasing N/L ratio. For these reasons, ESAE mice were more active and less stressed or fearful than TAA-supplemented mice.

The information of the current study uncovered that oral administration of TAA causes a critical rise in absolute WBC count, neutrophil count, N/L proportion, and a huge reduction in platelet count. **De Jong et al., (2002)** mentioned that an increase in the N/L ratio acts as an indicator of mild to moderate stress. This information concurred with **Abbasi et al., (2013)**, who announced that intense TAA poisonousness in rodents prompted leukocytosis, neutrophilia, and thrombocytopenia, which may be because of tissue rot or localized necrosis coming about because of openness to the deadly portion of TAA. While the oral administration of ESAE shows a critical increase in neutrophil count and platelet count while, it significantly decreases N/L proportion.

The current study's data agreed with **Ahmed (2014)** who reported that oral administration of *Eruca sativa* oil to female albino rats for 1 week stimulates leukocytosis. This huge neutrophilia could mirror its inclusion in the inflammatory process by forming different receptive oxygen species (ROS) or because of the fast arrival of youthful cells from the bone marrow (**Doi et al., 1991**).

The current study data revealed that TAA oral administration significantly decreases the total plasma protein and globulin concentration and increases the A/G ratio. On the other hand, ESAE significantly increases the total plasma protein and globulin concentration while decreasing the A/G ratio. As per **El-Missiry and El-Gindy, (2000)**, the capacity of *Eruca sativa* oil to invigorate the recovery of hepatic tissue is the primary driver of expanded protein production in harmed liver cells and improvement in the physiological function of the liver cells. These data disagreed with (**Ahmed, 2014**), who uncovered that the *Eruca sativa* oil treatment on profenofos enhanced female albino rodents had a non-significant impact on total plasma protein and albumin concentration despite recording, in some cases, a high percentage of differences, which may be due to higher standard errors. Also, the present results were dissimilar to those of **El-Nameary et al., (2016)** who observed that the prolonged addition of *Eruca sativa* seed meal in the diet of male rabbits has no significant effect on plasma protein, albumin, globulin, or A/G ratio.

The current results reported that TAA administration shows significant elevation of plasma biochemical parameters like AST and ALT, which are primary markers for a liver function test. The elevation

of these biomarkers may be due to systemic damage of the hepatic cells by reactive oxygen species released by the TAA. While the treatment with ESAE significantly reduces the plasma AST and ALT. The current study results were in concurrence with **Shyamal et al., (2010)** who inferred that the organization of TAA at portions of 50 mg/kg/i.p. on the other hand, like clockwork for 21 days brings about hepatic harm in animals. The component behind its poisonousness is related to its harmful metabolite (s-oxide). It slows down the development of RNA from the core to the cytoplasm, which might cause film injury (**Anbarasu et al., 2012**). The liver cell populace's injury is because of the expanded creation of TAA metabolites, including sulfines and sulfene (**Chanda and Mehendale, 1994**).

These results agreed with **El-Nattat and El-Kady (2007)**, who mentioned that the administration of *Eruca sativa* resulted in promoting ALT and AST activities in male rabbits, which may possibly be because of the high sulfur content in *Eruca sativa* that removes body wastes, clears congestion such as sinusitis and supports the liver and immune function. *Eruca sativa* leaves and seeds have a strong free radical scavenging antioxidant activity and protect the body cells from damage caused by oxidation through maintaining or raising the levels of antioxidant molecules and antioxidant enzymes (**Alam et al., 2007**). Additionally, these outcomes were as per **Alqasoumi (2010)** who detailed that *Eruca sativa* extract safeguards the liver against CCl<sub>4</sub> initiated hepatic injury through its powerful cancer prevention agent movement in rodents.

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#### **CONCLUSION**

It is concluded that ESAE at 1% daily oral dose possesses immunostimulant and hepatoprotective activity through inhibiting the reactive oxygen species produced by the administration of TAA. Therefore, ESAE improves mouse activity and reduces the anxiety induced by TAA. Further examinations are expected to portray its dynamic standards and explain its hepatoprotective movement's instrument.

**Declaration of Conflicting Interests:**

The authors revealed that there is no potential conflicts of interest.

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