Effect of Dietary Supplementation of Lemon Balm (*Melissa officinalis L.*) on Acute Kidney Injury in Rats

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Abstract

This study attempted to examine the impact of underlying processes of lemon balm leaves (*Melissa officinalis L.*) on rats with acute kidney injury (AKI) provoked by glycerol. Thirty male albino rats were utilized and weighed 170 ± 5g and divided into five groups (six rats of each). Group 1 was maintained on the standard basal diet and served as a negative control group. Groups 2, 3, 4 and 5 were administered glycerol (10 ml/kg b wt., fifty percent v/v in sterile saline, i.p.) to induce AKI. Group 2 was fed on the basal diet and kept as a positive conrol group, while the others three groups fed on the supplemented basal diet with 5, 10 and 15% lemon balm leaves, respectively. The findings indicated that treated AKI groups with lemon balm leaves (5%, 10%, and 15%) have variations in BWG, feed efficiency ratio & feed intake. As well, there is a significant decrease ($P < 0.05$) in liver and kidneys weights of treated AKI groups with lemon balm leaves, compared with that of the positive control group. The positive control group had significantly higher levels of liver functions (ALT, AST & ALP), lipid profile (cholesterol, triglycerides, LDL-c, VLDL-c), kidney functions (uric acid, blood urea nitrogen & creatinine), malondialdehyde and glutathione and decrease in HDL-c, comparison to the treated AKI groups with the different levels of lemon balm leaves. Accordingly, lemon balm was suggested in this investigation to affective in acute renal disorders.

Keywords: Lemon Balm, Antioxidant Enzymes, Acute Kidney Injury, Liver Functions.

Introductions

Acute kidney injury (AKI) is a common and critical issue that affects hospitalized cases. It is associated with elevated rates of morbidity and mortality, extended hospital stays, expensive medical costs **(Chertow *et al.*, 2005)**. A rapid decline in kidney function is the hallmark of AKI **(Kellum *et al.*, 2012).** Since, the fact that the criteria for defining & staging AKI defined by international consensus depends only on variations in serum creatinine (SCr), urine output (oliguria) & glomerular filtration rate (GFR), the timely identification, differential diagnosis, and management of AKI keeps presenting difficulties. **(Kalum and Lameire, 2018 & Selby, 2019).** According to **Moore *et al.*, (2018)**, AKI is a complex clinical case with a significant Rate of mortality. Even with the availability of cutting-edge renal replacement technologies, AKI patients still have very dismal outcomes. The kidneys of AKI patients lose their ability to excrete, which causes oliguria within a few hours or days. AKI is typically identified in people who are very sick and admitted to hospital intensive care units. While the most of AKI patients show signs of kidney functioning recovery, many more develop renal impairment or become dependent on dialysis (**Lameire *et al.*, 2008**). A sudden decline in glomerular filtration function is the hallmark of the illness known as acute kidney injury (AKI), or more precisely, a set of disorders. AKI is a widespread ailment that affects people worldwide, irrespective of their financial standing. The syndrome is linked to significant morbidity, mortality, and expensive treatment. Since end-stage renal disease (ESRD) &CKD emerged from an AKI episode, and since AKI is becoming more common, its effects on long-term health and expenses are significantly more significant than previously recognized **(Mehta *et al.*,2015).**

Lemon balm (Melissa officinalis) is one of the old herbal remedies from the Lamiaceae family used in treatment and thrives in Asia, Europe, and North America. It was first used as a medicinal plant in Mediterranean nations as antiviral, antimicrobial, antibacterial, anti-inflammatory, antioxidant and antispasmodic qualities in both its extracts and essential oil (EO). As well, it is advantageous against a variety of ailments., including Alzheimer's, cancer & HIV-1. The antioxidant and antibacterial properties of lemon balm can be assigning to its abundance of phenolic chemicals, including thymol and carvacrol (**Sharifi-Rad *et al.*, 2021; Sohrab *et al.*, 2021 & Zam *et al.*, 2022**). One of the most ancient and well-known herbaceous fragrant herbs, *M. officinalis*, has been applied to use in numerous methods. including compresses, ointments, and oil and aqueous extracts. The plant's sedative, antioxidant, anti-anxiety, neuroprotective & hypnotic qualities are made possible by a number of active components. The plant extract protects the liver, lowers cholesterol profiles, and modifies thyroid hormone activity, among other metabolic interventions. The plant has a distinct quality due to the presence of potent antioxidants, in addition to cytotoxic and anti-mutagenic propertiesexclusive capability of inhibiting free radical production **(Zarei *et al.*, 2015).** *M. officinalis or* Lemon balm., is employed in old treatment for sedation and memory enhancement. If taken at the right concentration, it also has other health benefits, but as of now, no thorough compilation has been completed **(Swiader *et al.*, 2019).**

According to **Vanti *et al.*, (2020),** essential oils from *M. officinalis* consist of specific volatile chemical constituents that are hydrophobic, concentrated & susceptible to changes in temperature, moisture, light, and oxygen. According to **Aziz *et al.*, (2018),** there are several methods for extracting it from plants, including solvent extraction, distillation, using a resin binder, absolute

extraction, cold pressing. Essential oil (EO) is utilized for its odors or aromas in the perfume and cosmetics industries as well as in the food business**.** EOs has a wide range of therapeutic properties. This means that the best way to classify or organize Eos is according to the plants they come from. Saying "essential oils" of chamomile, peppermint, tea tree, lavender, or *M. officinalis* is more helpful and distinctive **(Lee *et al*., 2012&Peterfalvi *et al*., 2019)**.

The most significant components of plants are flavonoids like monoterpene derivatives, luteolin, and apigenin; sesquiterpenes, including germacrene and beta-caryophyllene; triterpenes, including oleanolic and ursolic acid; volatile oil; tannins; and phenolic compounds, including, caffeic acid, rosmarinic acid, metrilic acid and cholinergic acid; are all present **(Rasmussen, 2011).** The presented research attempts to investigate the potential health advantages of *M. officinalis*, with a focus on the kidney functions and some of specific biochemical markers.

Materials and Methods

Materials:

Lemon balm leaves: The lemon balm leaves were acquired from (Shana company in Shepen Elkom, Monofia, Egypt). The Department of Medicinal Plants at the National Research Center were classified the plants as following.

Rank	**Scientific Name**
Kingdom	*Plantae*
Division	*Tracheophyta*
Subdivision	*Speramtophyta*
Class	*Magnoliopsida*
Superorder	*Asteranae*
Order	*Lamiales*
Family	*Lamiaceae*
Genus	*Melissa*
Species	*Melissa officinalis* L

Rats: Thirty adult male albino rats of the Sprague-Dawley strain, weighed 170 ± 5g were obtained from Health Research Institute Agricultural Research Center in Giza, Egypt.

Chemicals: Glycerol was obtained from Sigma Company for chemicals and pharmaceutical industries in Cairo, Egypt. The analytical reagents and chemicals that were used in the experiments were obtained from El-Gomhoria company for trading drugs, chemicals, and medical instruments, an organization based in Cairo, Egypt.

Methods:

Chemical composition: The moisture content of plant leaves was determined by dehydrating them until to a constant weight at 105 °C according to the method of **Anonymus, (1990)**. While ash, the total protein and the cellulose (crude fiber) content of plant leaves were determined as described by **Horwitz and Latimer, (2005); Latimer, (2016)** & **Brendel *et al*., (2000),** respectively**.** The total carotenoids, phenolic as Folin-Ciocalteu Reagent (FCR), and the flavonoid content were determined according to the methods of **Corte-Real *et al*., (2017); Singleton *et al*., (1999) & Biju *et al*. (2014**), respectively.

Experimental study: The experiment was performed in Animal Health Research Institute Agricultural Research Center. Rats were housed in hygienic conditions, kept for one week of adaptation, a standard diet was provided **(Reeves *et al.*, 1993).** Following the seven days of adaptation, rats were randomly divided into five groups as following:

Group 1: Feed on the basal diet and serving as a healthy control group.

Group 2: Feed on the basal diet & intraperitoneal injected with10 ml/kg bwt. v/v of glycerol / saline solution according to **Zager, (2015).**

Group 3: Feed on the supplemented basal diet with 5% lemon balm leaves & intraperitoneal injected with10 ml/kg b wt. v/v of glycerol / saline solution.

Group 4: Feed on the supplemented basal diet with 10% lemon balm leaves & intraperitoneal injected with10 ml/ kg b wt. v/v of glycerol / saline solution.

Group G5: Feed on the supplemented basal diet with 15% lemon balm leaves & intraperitoneal injected with10 ml/ kg b wt. v/v of glycerol / saline solution.

Daily food intake (FI), body weight gain, and feed efficiency ratio (FER) were calculated using the methods of **Chapman *et al.*, (1959)**. After the six-week experiment was over, the animals were anesthesia with diethyl ether. Blood samples were taken, held for thirty minutes, and then centrifugation for 15 minutes at a speed of 3000 rpm to serum separation, which was then gathered and stored in sterile containers at -20°C until used for biochemical analysis (**Helal *et al.*, 2012**).

Biochemical Analysis:

- **Assay of Liver Enzymes:** Activity of aspartate aminotransferase (AST) & alanine aminotransferase (ALT) and alkaline phosphatase (ALP) enzymes were assessed according to the methods of **Breuer, (1996)** & **Roy, (1970),** respectively.
- **Assay of Kidney Functions**: serum urea nitrogen, uric acid and creatinine were determined using the methods of **Patton and Crouch, (1977)** & **Jaffe, (1980)**, respectively.
- **Assay of Serum Lipids Profile:** Serum triglycerides (TG) were measured using **Fossati and Prencie, (1982)** methodology. **Henry *et al.*, (1974)** protocol was used to measure serum total cholesterol (TC). The concentration

of HDL-cholesterol in the serum was determined according to **Burstein, (1970)**. The formula of **Friedewald *et al.*, (1972)** was used to calculate the low-density lipoprotein cholesterol LDL-cholesterol and VLDL-cholesterol concentrations in serum as follows

LDL-c (mg/dl) =TC-(HDL-c + VLDL-c).
VLDL-c (mg/dl) = (Triglycerides /5)

- **Determination of Serum Malondialdehyde and Antioxidant Enzyme**: serum malondialdehyde (MDA) and activity of glutathione (GSH) enzyme were measured according to **Sinha, (1972)** & **Draper and Hadly, (1990),** respectively.

-Statistical Analysis: The standard deviation of each value was included in the mean. The statistical analysis of the data was conducted utilizing the computerized SPSS (Statistical Software, Cary, NC; Sigmastat. SPSS Program). The analyses of variance (ANOVA) were employed to examine the impacts of various interventions. Duncan's multiple range test was utilized to determine significance between groups, and a degree of significance of $p < 0.05$ was employed to denote this**. (Snedecor and Cochran, 1967).**

Results and Discussion

Table (1) displays the total contents of protein (11.43g), cellulose (25.34g,), ash (8.21g), moisture (8.55g), total carotenoids (2.3 µg), total flavonoid (10.31 mg of flavonoid quercetin equivalents) and phenolic (75.33 mg gallic acid equivalent) of lemon balm leaves were measured. As reported by **Moradi *et al.*, (2016)** found that methanolic extract of lemon balm leaves has 227.6 mg / GAE of phenolic content. A different investigation found that the total phenolic content was 54.9±2.14 mg GAE (**Spiridon *et al.*, 2011**). The average value for

the total phenolic in methanolic leaf extracts was 71.02 mg GAE (**Hassan *et al.*, 2019**). Also, the findings were corroborated by **Tusevski *et al.*, (2014)**, who revealed that the mean value of total phenolic content in lemon balm was 70.86 mg GAE. In a study by, **Spiridon *et al.*, (2011)** determined the total phenol content of lemon balm was 25.8±6.26 mg GAE.

Flavonoid compounds in the plant inhibit the production of prostaglandins & inflammatory cytokines through the inhibition of cyclooxygenase as a response to the inflammatory stimulation. thereby preserving and maintaining catecholamines. Additionally, this can be utilized to enhance their anti-inflammatory responses (**Vaez *et al.*, 2011**). The extract of *M. officials* is believed to include rosmarinic acid, flavonoids & terpenoids, which have anti-nociceptive and anti-inflammatory properties. It's likely that flavonoids work more efficiently via promoting prostaglandin production. Flavonoids exert their analgesic effect by modulating the opioidergic pathway **(Miladi-Gorgi *et al.*, 2005 & Anjaneyulu and Chopra, 2003)**.

Medicinal herbs have a diverse array of bioactive chemicals, as demonstrated by recent developments in pharmacognostics. Among them, phytochemical investigations have found that lemon balm contains a variety of volatile (like neral & geranial), polyphenolic (like luteolin, rosmarinic acid and naringin), and terpenoids (like ursolic acid) chemicals **(Shakeri *et al.*, 2016).** Similar to this, a variety of terpenoids (like taraxacinic acids), alkaloids (like taraxacine and taraxafolin), and polyphenolic compounds (like chicoric acid, chlorogenic acid, luteolin & isorhamnetin) have been separated to the aerial parts of dandelion **(Schutz *et al.*, 2006).** Polyphenols are responsible for much of antioxidant and anti-inflammatory properties found in plants. A combination of lemon balm and dandelion extracts effectively solubilizes two such chemicals, namely chicoric acid & rosmarinic acid which contain rosmarinic acid at a

concentration of 34.07 ± 0.55 mg/g and chicoric acid at 2.26 ± 0.01 milligram/gram, both of which are found in lemon balm and dandelion. **(Choi *et al*., 2020).**

Pereira *et al*., (2009) reported that phenolic components in the plant extract were also shown to possess antioxidant properties. are largely related to rutin, quercetin, garlic acid & quercetin. Quercetin has the strongest antioxidant qualities among the chemicals, followed by rutin, garlic acid, and quercetin, in that order. Nitric acid (30.44%), isopolcule (22.02%), citral (27.03%), oxide carolyn (1.24%), cariophiline (2.29%) and citronella (1.06%) are all present in the *M. officinalis* oil extract that is made from the plant's leaves. Its distinct and potent analgesic & anti-inflammatory properties have been demonstrated in animal models in contrast to those of anti-inflammatory& analgesic standard medication (indomethacin) **(Bounihi *et al*., 2013).** According to a phytochemical analysis of *M. officinalis*, garlic acid &rosmarinic acid are the phenolic compounds with lowest amounts& the highest, respectively **(Arceusz and Wesolowski, 2013).** The flavonoid constituents, including quercetin, luteolin, and rhamnocitrin, among others, enhanced the medicinal utility of the species. In addition to enhancing memory and mood (**Dehbani *et al*., 2019**& **Kennedy *et al*., 2002),** The administration of polyphenols in elevated concentrations was employed to treat specific digestive and gastrointestinal disorders.

Table (1): Chemical composition of lemon balm leaves.

Parameters	Contents
Protein	11.43gm
Cellulose	25.34gm
Ash	8.21gm
Moisture	8.55gm
total carotenoids	2.3 µg
Total phenolic	75.33 (mg GAE)
Total flavonoid	10.31 (mg QE)

The results shown in Table (1) showed that the percentage of body weight growth in the positive control group was significantly lower ($p < 0.05$) than in the negative control group (143.60±2.70 VS 159.40±1.81).Reduced palatability of the feed mix may be the cause of the observed decreases in body weight growth, feed efficiency, feed intake, and feed efficiency ratio in rats induction with glycerol (+ve). There was a substantial ($P < 0.05$) increase in the percent of BWG in tested groups comparing to the positive control group. By comparison, the negative control group eat more food than the positive control group. In all treated groups, there were non-significant variations in FER compared to healthy group. The higher body weight gain and enhanced concentrations of *Melissa officinalis* groups may be related to the rats' increased sensory, tasting, and palatability of the meal.

El-Gamel, (2021) found that when treated rats fed diets containing 5%, 7.5% and 10% melissa powder, the differences in FI, BWG, and FER between the rats in both the positive and negative control groups were not statistically significant. Our findings support the findings of **(Sief *et al*., 2015),** who found that rats treated with the herb melissa officinalis (MO) exhibited a statistically significant increase in feed intake and body weight.

Table (2): Effect of lemon balm leaves on body weight gain , feed intake and feed efficiency ratio of rats with acute kidney injury.

Parameters / Groups	Bodyweight gain (%)	Feed intake (g/day)	Feed efficiency ratio
Control(-Ve)	159.40±0.81[a]	15.08±0.30[a]	10.57±0.16[b]
Control(+Ve)	143.60±1.20[d]	11.62±0.38[d]	9.45±0.38[a]
Lemon Balm Leaves (5%)	156.00±2.21[b]	14.40±0.07[b]	10.79±0.19[b]
Lemon Balm Leaves (10%)	155.60±1.16[b,c]	13.52±0.42[c]	11.52±0.21[b]
Lemon Balm Leaves (15%)	150.00±2.12[c]	13.16±0.31[c]	11.44±0.07[b]

*Values are expressed as means ±SE.
*Values at the same column with different letters are significantly different at P<0.05

Table (3) illustrates outcome of changes in the organs relative weights. The liver and kidney have mean relative weights were considerably lower in positive control group of rats induction with glycerol (AKI) than in negative group. Compared to the positive control group, treating the AKI groups with the three different amounts of lemon balm leaves resulted in a substantial rise ($p \leq 0.05$). The information in this table showed that there were not substantial variations in kidney and liver organ weight in all test groups.

Table (3): Effect of lemon balm leaves on liver and kidney relative weight of rats with acute kidney injury.

Parameters / Groups	Liver	Kidney
	%	
Control(-Ve)	4.16±0.12[a]	0.63±0.04[a]
Control(+Ve)	2.77±0.11[b]	0.50±0.11[b]
Lemon Balm Leaves (5%)	4.49±0.10[a]	0.95±0.03[a]
Lemon Balm Leaves (10%)	4.35±0.08[a]	0.79±0.11[a]
Lemon Balm Leaves (15%)	4.25±0.07[a.]	0.64±0.13[a]

*Values are expressed as means ±SE.
*Values at the same column with different letters are significantly different at P<0.05

The impact of lemon balm on alanine aminotransferase (ALT) was demonstrated by the data in table (4). The serum ALT level was 66.40± 2.51 (U/L) the value was considerably greater in positive control group compared to negative control group. The findings demonstrated that, serum ALT levels decreased in rats fed on diet supplemented with lemon balm at any dose, in contrast to positive control group.

Lemon balm leaves has an effect on aspartate aminotransferase (AST) serum activity, as shown by table 4 data. The results demonstrated a considerable increase in AST activity with a mean value of 111.20±3.42 (U/L) in rats receiving glycerol (positive control group) compare to negative control group 38.80±2.04 (U/L). In contrast to (+ve) control groups, rats given a basal diet with three different levels of lemon balm show a substantial decrease in serum AST activity.

Based on the data, it was observed that (+ve) control group exhibited the most significantly elevated serum alkaline phosphatase (ALP) level of 855.60±1.14 (U/L). The results demonstrated that the serum ALP concentration of rats fed any concentration of lemon balm was less than that of (+ve) control group.

When *M. officinalis* was given to a hypercholesterolemic group, **Zarei *et al*., (2014)** report a decrease in the levels of liver enzymes. Hepatic dysfunction induced by hepatic fat accumulation manifests as increased concentrations of hepatic enzymes. especially alanine aminotransferase **(Murray *et al*., 2012)**. Conversely, as lipid levels rise due to liver hyperlipidemia, free radicals (FRs) are released **(Nazari *et al*., 2005).** Our findings are consistent with those of **Elsadek and Habib, (2018),** who suggested that a possible explanation for the significant decreased in serum levels of aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH) and gamma-

glutamyl transferase (GGT) in rats given lemon balm was a reduction in liver cell leakage. This implies that the hepatic damage caused by the lemon balm leaves was repaired, and the cellular permeability was restored, lessening the oxytetracycline's harmful effects on hepatic cells. It appears that lemon balm's potent antioxidant qualities are the reason behind its renowned ability to lower liver enzyme levels. One of the most significant classes of antioxidant agents found in this plant are phenolic compounds (**Zarei *et al*., 2014).** Phenolic compounds that are detected in the extract of the phenolic compounds present in the extract of *M. officinalis* possess antioxidant properties, enabling them to effectively eliminate free radicals (FR). By inhibiting the cytochrome system, and flavonoids, which boost the capacity of antioxidant enzymes and shield cells against glutathione depletion, potentially explains the extract's hepatoprotective effect **(Chung *et al*., 2010)**. In a different investigation, **Bolkent *et al*., (2005)** looked at how *M. officinalis* extract affected the livers of hyperlipidemic rats. They found that while glutathione levels increased, liver enzymes and cholesterol decreased.

It is well known that *M. officinalis* extracts potent antioxidant qualities contribute to its ability to lower liver enzyme levels. Phenolic chemicals, which rank among the most significant antioxidant agents, are present in this plant. These substances, particularly flavonoids exhibit cytochrome system inhibitory properties, which protects the liver from damage brought on by free radicals. By boosting the activity of glutathione reductase, oxidase, the antioxidant enzymes ,catalase, flavonoids can also shield cells against glutathione depletion **(Zarei *et al*., 2014).**

Hepatocyte cell membranes are harmed by free radicals. The breakdown of the hepatocyte membrane causes a heightened level of activity of hepatic enzymes, which causes the production of these enzymes that are typically found

in the cytoplasm of cells. The kind and extent of liver injury are explained by a heightened level of activity of these enzymes. The reduction in hepatic enzyme activity in the treatment groups receiving *M. officinalis* extract is expected given the features of this herb, which include its anti-inflammatory and antioxidant properties additionally with its capability of inhibiting the development of free radicals **(Shariati and Zarei, 2006).** The present research revealed that *M. officinalis* extract administration be able to preserve liver cells as a result of flavonoids.

Table (4): Effect of lemon balm leaves on serum liver enzymes of rats with acute kidney injury.

Parameters / Groups	ALT	AST	ALP
	(U/L)		
Control(-Ve)	23.60±1.12^{e}	38.80±0.91^{d}	802.40±0.98^{b}
Control(+Ve)	66.40±1.12^{a}	111.20±1.53^{a}	855.60±0.51^{a}
Lemon Balm Leaves (5%)	51.20±0.73^{b}	63.20±0.73^{b}	793.00±0.89^{c}
Lemon Balm Leaves (10%)	47.00±0.54^{c}	43.40±0.98^{c}	783.60±1.36^{d}
Lemon Balm Leaves (15%)	32.20±1.49^{d}	44.60±0.24^{c}	772.20±1.02^{e}

*Values are expressed as means ±SE.
*Values at the same column with different letters are significantly different at P<0.05

Table (5) demonstrates the impact of lemon balm leaves on renal function in rats with acute kidney injury (serum concentrations of urea, creatinine and U.A). The administration of glycerol to rats resulted in a major increase in serum urea concentration, with mean value of 46.20±1.30 (mg/dL), compare to the (–ve) control group 32.40±5.51 (mg/dL). However, the serum urea concentrations of rats induction with glycerol and fed on diet supplemented with lemon balm at each of the three concentrations decreased significantly compared to positive control group.

The uric acid levels in (+ve) control group were found to be the highest in

compared to(–ve) control group, with a mean values of 6.32±0.57 mg/dL and 3.62±0.27 mg/dL, respectively. Additionally, the serum uric acid concentration of the groups (3,4 and 5) fed on lemon balm leaves decreased significantly compared to positive control group.

Additionally, as shown in Table (4) the positive control group that administered glycerol exhibited a higher concentration of creatinine. In comparison to (–ve) control group 0.70±0.01 (mg/dL), positive control group exhibited a substantial increase in the mean value of serum creatinine, which reached 1.80±0.03 (mg/dL) on average. Comparatively, serum creatinine levels in AKI groups fed on diet supplemented with the lemon balm leaves decreased substantially when compared to positive control groups.

According to **Tanaka *et al*., (2022)** the kidney protection effect of lemon balm methanol extract is demonstrated through a substantial reduction in bilirubin(BUN) and creatinine concentrations. With the exception of the maximum dose of lemon balm methanol extract (1,200 mg/kg B.W.), no microscopic enhancement of the kidney was observed at any other dosage. Additionally, research by **Worotikan *et al*., (2017)** has shown that a 300 mg/kg body weight dose of lemon balm ethanol extract protects the kidneys of wistar rats induced with diabetic alloxan. In addition to adenine, kidney dysfunction was induced by a significant rise in serum urea and creatinine levels . However, renal function indicators returned to normal values following treatment with lemon balm leaves, which can be attributable to the plant's distinctive property of bioactive and effective antioxidants, particularly those with the capacity to obstruct the generation of free radicals. The findings presented here are consistent with previous research**. Namjoo *et al*., (2013)** demonstrated a significant decrease in serum urea and creatinine levels following treatment by lemon balm extract.

Table (5): Effect of lemon balm leaves on kidney functions of rats with acute kidney injury.

Parameters / Groups	Urea	Uric acid	Creatinine
	mg/dl		
Control(-Ve)	32.40±1.21^{d}	3.62±0.12^{c}	0.70±0.01^{c}
Control(+Ve)	46.20±0.58^{a}	6.32±0.25^{a}	1.80±0.03^{a}
Lemon Balm Leaves (5%)	38.60±0.87^{b}	4.42±0.13^{b}	1.01±0.05^{a}
Lemon Balm Leaves (10%)	37.20±1.15^{b,c}	4.24±0.07^{b}	0.73±0.01^{b}
Lemon Balm Leaves (15%)	36.00±0.54^{c}	3.96±0.15^{c}	0.69±0.04^{c}

*Values are expressed as means ±SE.
*Values at the same column with different letters are significantly different at $P<0.05$

As shown in Table (6) the positive control group has significantly increased levels of TG, TC, VLDL-C and LDL-C ($P\leq0.05$). In contrast, HDL-C concentrations were considerably decreased compared to negative control group. The incorporation of lemon balm in the diet resulted in decreased in the concentrations of lipid profiles in the serum, with the exception of HDL-C, which increased significantly compared to the positive control group. Best results were in groups of rats with AKI that were fed on lemon balm leaves compared to positive control group. In the treatment or prevention of acute renal disease, these modifications illustrate the significance of a diet rich in *M. officinalis*.

Daily consumption of *M. officinalis* tea can reduce TG and lipid metabolism in humans **(Jun *et al.*, 2012).** In addition, the potential effect of *M. officinalis* can prevent hypercholesterolemia, hyperlipidemia and lipid peroxidation in rat liver **(Bolkent *et al.*, 2005)**. In hypercholesterolemic rats, the research of **Changizi-Ashtiani *et al.*, (2013)** showed that lemon balm reduced cholesterol, LDL and triglycerides. These effects might be associated with the antioxidant properties of *M. officinalis* which increase the levels of thyroid hormones or the

plant contains quercetin compounds, which exhibit inhibitory properties towards lipid peroxidation **(Dolatabadi *et al*., 2018).** A research by **Jun *et al*., (2012)** The potential mechanism by which *M. officinalis* extract could decrease plasma triglyceride levels was ascribed to the presence of quercetin, a compound identified in the plant., that might influence the incidence of lipid peroxidation in the oral cavity.

According to recent studies, the metabolic properties of *M. officinalis* essential oil are physiologically substantial. Essential chemicals in vegetable oils contain terpenoids that reduce cholesterol by inhibiting hepatic biosynthesis and formation of biliary cholesterol nuclei **(Chung *et al*., 2008).** Cholesterol levels can be improved with daily and consistent consumption of *M. officinalis* tea. triglyceride and other metabolic factors in humans Furthermore, *M. officinalis* has the ability to prevent excess cholesterol, increase blood pressure cholesterol in the liver of over fat rats is decreased, reducing lipid peroxidation **(Bolkent *et al*., 2005).** According to the available evidence, medicinal oils, including *M. officinalis* oil, possess a variety of pharmacological effects that are primarily attributed to volatile terpenoids, including cineole, caffeic acid, and geranial. **(Jun *et al*., 2012).**

Research on *M. officinalis* has also revealed the presence of phenolic alkaloids, which are among the substances that might prevent the manufacture of cholesterol **(Pereira *et al*., 2009& Ashtiyani *et al*., 2011).** The findings of this investigation are consistent with those of **Bolkent *et al*., (2005)** The researchers, along with their collaborators, provided evidence that the administration of *M. officinalis* extract resulted in reductions in lipid peroxidation (LPO) levels in hepatic tissue, total cholesterol and total lipid present in serum level.

Table (6): Effect of lemon balm leaves on serum lipid profile of rats with acute kidney injury.

Parameters / Groups	TC	TG	HDL-c	LDL-c	VLDL-c
	(mg/dl)				
Control(-Ve)	92.76±0.87^{e}	96.20±1.24^{e}	34.80±0.37^{c}	38.80±0.92^{b}	19.24±0.48^{e}
Control(+Ve)	120.76±1.17^{a}	186.20±4.31^{a}	30.80±0.37^{d}	33.72±1.45^{c}	37.24±0.86^{a}
Lemon Balm Leaves (5%)	104.54±1.03^{d}	174.00±1.22^{b}	40.80±0.37^{a}	28.94±1.13^{d}	34.80±0.24^{b}
Lemon Balm Leaves (10%)	111.84±0.60^{c}	164.20±0.97^{c}	40.40±0.51^{a}	38.60±0.99^{b}	32.84±0.19^{c}
Lemon Balm Leaves (15%)	107.48±1.13^{b}	141.60±2.13^{d}	37.60±0.51^{b}	41.36±0.81^{a}	28.32±0.42^{d}

*Values are expressed as means ±SE

*Values at the same column with different letters are significantly different at P<0.05

The data presented in Table (7) indicates a statistically significant increase in the mean value of malondialdehyde activity in (+ve) control group compared to the normal group (P>0.05). The average glutathione activity of rats fed on treated diet was significantly increased (P>0.05) than that of positive control group. In rats exposed to *M. officinalis* , the average malondialdehyde activity decreased substantially (P>0.05) as a result of nephrotoxicity, compared to positive control group. Additionally, when rats were fed *M. officinalis*, the average levels of glutathione heightened activity substantially across all groups that contrast to positive control group (P>0.05). An evaluation of the *M. officinalis* sample revealed that it enhanced the activity of malondialdehyde and glutathione.

M. officinalis has antioxidant properties and is as a result of the existence of rosmarinic acid and benzodioxoli .The antioxidant properties of these compounds in the extract are ten times more potent than for both vitamins B and C. Thus pharmaceutical products such as melissa root toxin officinalis Vitamin C to facilitate the effects **(Ghayoor *et al.*, 2010)**. In addition, the

extract also contains compounds like acidic carnosic acid, linoleic acid and urosolic acid, each possessing antioxidant properties. Additionally, compounds derived from *M. officinalis* inhibit the enzyme acetylcholinesterase (AChE) and acetylcholine binding and can improve cognitive functions such as memory **(Rostami *et al*., 2010).**

Extensive testing was conducted on the antioxidant properties of *M. officinalis* leaves. The outcomes demonstrated that this essential oil possesses anti-inflammatory properties, in addition to its conventional use in treating a range of conditions associated with pain and inflammation. **(Bounihi *et al*., 2013).** Recent work has shown that *M. tuberculosis. M. Officinalis* extract showed anti-inflammatory effects by means of L-arginine nitric oxide pathway interaction with muscarinic and nicotinic receptors which consisted of terpenoids and flavonoids **(Miladi Gorgi *et al*., 2005).** flavonoids have been identified as inhibitors of several inflammation-related enzymes, including lipoxygenase, monooxygenase and cyclooxygenase **(Petersen and Simmonds, 2003).**

Table (7): Effect of lemon balm leaves on serum malondialdehyde and glutathione of rats with acute kidney injury.

Parameters / Groups	MDA (nmol/min/mg protein)	GSH (U/mg protein)
Control(-Ve)	66.00±0.83^{e}	3.74±0.08^{b}
Control(+Ve)	152.20±1.02^{a}	2.12±0.11^{c}
Lemon Balm Leaves (5%)	91.20±0.58^{b}	3.46±0.13^{b}
Lemon Balm Leaves (10%)	83.40±1.07^{c}	3.56±0.19^{b}
Lemon Balm Leaves (15%)	68.00±0.89^{d}	5.28±0.08^{a}

*Values are expressed as means ±SE.

*Values at the same column with different letters are significantly different at P<0.05

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تاثير المتناول من اوراق بلسم الليمون على الالتهاب الكلوى الحاد فى الفئران

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الملخص العربي

كان الهدف من هذا العمل هو دراسة تأثير اوراق بلسم الليمون على الفئران المصابه بالتهابات الكلى الحاده الناتجه عن الحقن بالجلسرين .تم استخدام ثلاثين فأر من ذكور الالبينو في هذد الدراسه وزنها ١٧٠ ± ٥ جرام .تم تقسيم الفئران إلى ٥ مجموعات (عدد = ٦ فئران) على النحو التالى المجموعة الأولى: تم تغذيتها على نظام غذائي عادي، واستخدمت كمجموعة ضابطه سالبه، المجموعة الثانية: تم تغذيتها على نظام غذائي أساسي بالإضافة إلى الجلسرين (١٠ مل /كجم من وزن الجسم، ٥٠٪ حجم/حجم في محلول ملحي معقم) لاحداث الاصابه بالقصور الكلوي الحاد وكانت مجموعة ضابطه موجبه، المجموعة ٣: تم حقنها بالجلسرين وتغذت على الغذاء الاساسى المضاف اليه ٥٪ بلسم الليمون . المجموعة الرابعة تم حقنها بالجلسرين وتغذت على الغذاء الاساسى المضاف اليه بلسم الليمون بنسبه ١٠ .%المجموعة ٥: تم حقنها بالجلسرين وتغذت على الغذاء الاساسى المضاف اليه ١٥٪ بلسم الليمون تم تقدير القيمه الغذائيه لاوراق بلسم الليمون .أظهرت النتائج أن جميع المجموعات المصابه بقصورحاد في الكلى وتناولت (٥٪, ١٠٪ ,١٥٪ بلسم الليمون) قد أحدثت زيادات متفاوتة في زيادة وزن الجسم وتناول الطعام ونسبة كفاءة التغذية .أظهرت النتائج وجود انخفاض معنوي ($P \geq 0.05$) بين المجموعات التي تم حقنها بالجلسرين والمحتوية على ٥٪، ١٠٪، ١٥٪ بلسم الليمون في أوزان الأعضاء الداخلية، مقارنة بالمجموعه الضابطه الموجبه .وظائف الكبد بما في ذلك (ALP, AST ,ALT)ووظائف الكلى (حمض اليوريك، ونيتروجين اليوريا، والكرياتينين)، ومستوى الدهون (الكوليسترول، والدهون الثلاثية، وLDL-c، وVLDL-c ، والمالوندياالدهيد زادت بشكل ملحوظ في المجموعه الظابطه الموجبه .مقارنه بالمجموعه الضابطه السالبه. بينما انخفض مستوى HDL-c والجلوتاثيون بشكل ملحوظ بالنسبه للمجموعه الضابطه الموجبه مقارنة بالمجموعه الضابطه السالبه .أظهرت جميع المجموعات التى تمت تغذيتها على ٥٪، ١٠٪ و١٥٪ من المليسانخفاضاً معنوياً في هذه المعايير باستثناءHDL ، مقارنة بالمجموعه الضابطه الموجبه .لذا أوصت هذه الدراسة بان بلسم الليمون فعال بالنسبه لامراض الكلى الحادة.

الكلمات المفتاحية :بلسم الليمون- الانزيمات المؤكسده –الالتهاب الكلوى الحاد-وظائف الكبد.