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The Effect Of Feeding With Molokhia (C. *Olitorius*) Leaves Powder On Lead Poisoning Rats.

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Abstract

Heavy metal poisoning, especially Lead poisoning has harmful effects to human health. Moreover, many searches found that there are some plants have anti-poisoning for metal. Thus, this research was carried out to study the effect of feeding Lead poisoning rats on 5, 10 and 15% molokiha (*Corchorus olitorius)* leaves powder for eight weeks on blood lead concentration, functions of kidney and liver and histological examination of liver, kidney and brain organs. The results showed that the molokiha leaves powder had high nutritional value; it had high contents of fiber, vitamins and total phenol. Moreover, as a result of feeding poisoning rat on molokiha leaves powder, the group feeding on 15% molokhia had the highest levels of feed intake, body weight gain and feed efficiency ratio compared to the positive control group and other groups feeding on molokiha. Moreover, the results indicated that the levels of blood lead concentration, liver enzymes and kidney function as urea nitrogen, uric acid and creatinine of Lead poisoning rats were decreased as a result of increase the levels of feeding on molokhia leaf powder. So, the group feeding on 15% molokiha had the lowest levels of all these analyses compared to the positive control group and other groups. Furthermore, histological examination of kidney, liver and brain organs of rats showed that the group feeding on molokhia 15% showed that the harmful of Lead poisoning in rats was decreased compared to the positive control group and other feeding

groups. However, it was insufficient to treat all changes in these organ tissues as a result of lead poisoning. Finally, the molokiha leave powder had high contents of compounds as fiber, total phenol and vitamins that had high antioxidant activity and antitumor for organ tissues of kidney, liver and brain. So, it can be utilized as an anti-poisoning for Lead poisoning.

Key words: Molokiha, Lead, poisoning, total phenol, vitamins, liver, kidney, brain, histological examination.

Introduction

Heavy metals become toxic when they are not metabolized by the body and gathered in the soft tissues. Heavy metals might come into the human body from water, food, air or absorption through the skin when they reach to humans from residential settings, industrial, pharmaceutical and agriculture **(Radostitis *et al.*, 1994).** Lead is considering toxicity, as one of the dangers problems of all the world; its particular, the certain and safe treatment was still generally anonymous **(Sheikh *et al.*, 2014).** In addition, the assimilation of lead happens more rapidly in children than in adults. Children, because of their childish behavior, are more prone to ingest and inhale dust polluted with Lead **(Landrigan, 2002).** Lead poisoning is a critical natural illness and its impacts on the human body are destruction. There is no capacity in the human body which isn't influenced by lead toxicity (**Wani *et al.*, 2015).** The harming of lead is different with the chemical substance type of the lead. Lead acetate derivation is extremely dissolvable and more poisonous than insoluble lead oxides, or soild lead sheeting **(Radostitis *et al.*, 1994).**

Lead exposure for the most part hails from the digestive system. It is bio-collected and is most concentrated in the liver. Lead over-burden of the lead causes of liver cells (starting the formation of tumors in the liver), and analogous to mercury, causes oxidation, inflammation, and increased blood LDL cholesterol levels. Liver cells die speedily and are substituted with fatty deposits. High levels of lead are united with a 3x-increment in liver deteriorates (high ALT levels) **(Cave *et al.*, 2010).** Lead acetate may be considered as a strong hepatotoxic and genotoxic

agent **(Mudipalli, 2007 and Haouas *et al.*, 2014).** Increasing blood lead levels are linked with mortality from chronic kidney diseases. This is clear in the range of blood lead level below10 μg/dl **(Bellinger 2011)**. Even when lead can reach and accumulate in almost every organ in the human body, the central nervous system is a specific goal of the harmful effects of metal. And the neurotoxic effects of lead are a major public health concern **(Verstraeten *et al.*, 2008).** Likewise, lead might have poisonous effects influence the intrinsic and the extrinsic induction of apoptosis pathway with prominent influence on brain tissue even at low dose (**Ahmed *et al.*, 2013).** The essential effect of lead on encephalic functions is deterioration the cell membranes. The nervous system is very sensitive to oxidative damage; since it is rich in oxidizable substrates that have a high a low antioxidant capacity and oxygen tension **(Sandhir *et al.*, 1994).**

The dietary plant plays an important character in healthcare management system and substantially, in the weight and feed efficiency due to its palatability and it is nutritional endowments (**Kunle *et al.*, 2017).** Molokiha can be used as a plant additive to other food and consumed by people. In general, it might be because of its health properties and its taste. There are needs for more research to decide the metabolite synthesis that happens in these types of plant, and in addition the development of new contents **(Ilhan et al., 2007).** Molokiha *C.olitorius* (Tiliaceae) is a yearly herb whose leaves and roots are utilized as natural drug and eaten as a vegetable by nearby individuals in East Malaysia, India, Egypt, and Philippines, South America and tropical Africa (**Zeghichi *et al.*, 2003 and Oyedeji and Bolarinwa, 2013).** *C. olitorius* (jute mallow) has diverse public names nalta jute, bush okra, ewedu, jute mallow, Jew's mallow, melokhia and monoheiya (**Fontem *et al.*, 2003 and Faith *et al.*, 2012).** Molokiha potential to enhance nutrition and health as a rich source of dietary phenolic and flavonoid, vitamin C, vitamin E, β-carotene, α-tocopherol, glutathione and phenols (antioxidants). The leaves also contain fatty acids, minerals, vitamins and polysaccharides for biological systems susceptible to free radicals-mediated reactions leading to oxidative stress in diseases **(Yokoyama *et al.*, 2014, Singh and Immanuel, 2014 and Adedosu *et al.*, 2015).** The antioxidant activity of phenolics depends on the redox properties of their hydroxyl groups, which enable them to do as agents

of reducing, metal chelators, hydrogen donors, and single oxygen quenchers **(Oboh and Rocha, 2007).** Also, it contains potassium, iron, vitamins and fiber than any other vegetable and hence it is called as the king of vegetables. It contains properties that help to boost the immune system and protects from cancer, high blood pressure, osteoporosis, premature aging, fatigue, and so on **(Ahmed and Nizam, 2008**).

Therefore, leaves from different Jute species have been used in folklore medicine for treating diabetes mellitus and hypertension. As reported for *Corchorus olitorius,* the mechanism might due to α-glucosidase and α-amylase inhibitory impact of some phenolic compounds as caffeic acid **(Ademiluyi *et al.,* 2014).** Also, using for cystitis, tumors, dysuria fever, pain, gonorrhea, strength and restore appetite **(Adediran *et al.,* 2015 and Ibrahim and Fagbohun, 2011)**. Molokiha leaves extracts against Lead induced toxic manifestation in blood, hepatic, renal, brain and cardiac tissues of Wistar rats (**Dewanjee *et al.,* 2013).** Most of the toxicities can be minimized through addition of *C. olitorius* leaves that contain antioxidants namely flavonoids, carotenoids and vitamin C. Therefore, *C.olitorius* can be enriched with the standard diet to the influenced people as prone zone **Hosen *et al.,* (2016).** The leaves of C olitorius have antitumors namely phytol and monogalactosyl-diacylglycerol (**Furumoto *et al.,* 2002 and Hosen *et al.,* 2016).**

For these reasons the aim of this research was studying the chemical composition of molokiha. Also, studying the effect of feeding poisoning rats on 5, 10, 15 and 20% molokiha leaves powder for 8 weeks on blood lead concentration, liver enzymes as (ALT and AST), kidney function as (urea nitrogen, creatinine and uric acid) and histological examination of kidney, liver and brain tissue compared to negative and positive control groups.

Materials And Methods

Materials

Fresh molokiha *(Corchorus olitorius)* were obtained from local markets in Beni Suef city government, Egypt.

Thirty five male albino rats of Sprague-Dawley strain weighting (200±10g) were obtained from the experimental animal house of Food Technology Research Institute, Agricultural Research Center, Giza, Egypt.

Casein, starch, corn oil, l-cystine, choline chloride, tert-butylhydroquinon,e all the vitamins, minerals, Lead (II) acetate trihydrate and kits for biochemical analysis (ALT and AST) were obtained from El-Gomhoryia, company Cairo, Egypt.

Methods

Preparation of molokiha leaves powder

The fresh molokiha leaves were washed and dried by sun drying. And it was grounded by an electric blender (Moulinex, LM207041 Super Blender, France) and was packed in low-density polyethylene bags until it was used for the required analysis.

Chemical analysis

Molokiha leaves powder was analyzed chemically for moisture, protein, fiber, ash, total sugars and oil (Fresh weight) contents were determined according to the methods described by **AOAC (2005).** Total polyphenol content (TPC) was estimated by the Folin-Ciocalteu method as described by **Ragaee *et al.*, (2006) and Dvorakova *et al.*, (2008).** Phenolic compounds were determined by HPLC according to **Goupy *et al.*, (1999)** Vitamin E (a-, b-, g-, and d-tocopherols) was determined according to **Pyka and Sliwiok, (2001).** Vitamin A were estimated according to **Noll, (1996).** Vitamin C was carried out according to **Romeu-Nadal *et al.*, (2006).** All these determinations were carried out in Agriculture Research Center, Cairo, Egypt**.**

Biological assay

Animal and experimental design:

Thirty five male Albino rats (Sprague-Dawley) weighing (200$\pm$ 10 g) g were kept under normal healthy conditions and fed on basal diet (experimental diets for adaptation) for seven days. Then, rats were divided randomly into five groups (n = 7). Basal diet, vitamin and mineral mixture compositions were prepared according to **Reeves *et al.*, (1993)** and was given in Table (1).

Twenty eight rats were fed on basal diets plus 0.005% Lead (II) acetate trihydrate [$Pb(CH_3COO)_2 \cdot 3H_2O$] powder for 4 weeks to toxic rats according to **Dewanjee *et al.*, (2013).** All diets were given in Table (1)**.**

Table 1: Composition of basal and poisoning rats feeding on molokhia leaves powder (g / 100 g).

Ingredients	Negative control group	Positive control group	Group feeding on 5% molokhia	Group feeding on 10% molokhia	Group feeding on 15% molokhia
Corn starch	62.0692	62.0692	57.0642	52.0642	47.0642
Molokiha leave powder	---	---	5	10	15
Casein(> 85% protein)	14	14	14	14	14
Lead (II) acetate trihydrate	--	0.005	0.005	0.005	0.005
Sucrose	10	10	10	10	10
Corn oil	4	4	4	4	4
Fiber	5	5	5	5	5
Mineral mix.	3.5	3.5	3.5	3.5	3.5
Vitamin mix.	1	1	1	1	1
L-cystine	0.18	0.18	0.18	0.18	0.18
Choline chloride	0.25	0.25	0.25	0.25	0.25
Tert-butylhydroquinone	0.0008	0.0008	0.0008	0.0008	0.0008
Total	100	100	100	100	100

During the experiment period (12 weeks) rats were kept separately in well aerated cages (stainless steel). The food intake (FI) and body weight gain (BWG) were recorded every week and at the end of the experiment. Feed efficiency ratio (FER) was calculated according to the method of **Chapman *et al*., (1959) by** using the following equation:

FER = Gain body weight (g) / Feed intake (g).

After poisoning period for four weeks and at the end of experimental period (12 weeks) rats were fasted overnight and anesthetized using diethyl ether and blood samples were taken from hepatic portal veins, the orbital venous plexuses by capillary tube into centrifuge tubes. Blood samples were centrifuged at 5000 r.p.m for 15 min to separate serum, and then kept in plastic vials at –20 ^{O}C until analysis. At the end of experiment liver, kidney and brain were removed and weighted to calculate its relative ratio to final body weight according to the following:

Relative organ weight = Organs weight (g) x 100 / Final body weight (g) according to the method of **Chapman *et al*., (1959)**.

Biochemical analysis of serum

The Lead concentration blood was determined according to **Parker and Absorpt, (1963)**

Serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined in serum samples using enzymatic colorimetric kits according to **Reitman and Frankel, (1957).**

Serum and urea nitrogen and uric acid were determined in the plasma according to the method described by **Patton and Crouch, (1977).** Serum creatinine concentration was determined as described by **Henry, (1974).**

Histological Examination:

Kidney, liver and brain were taken immediately after sacrificing the rats and immersed in 10% buffered neutral formalin solution, the fixed specimens then trimmed, washed and dehydrated in bedded, in paraffin cut into sections of 4-6 microns thickness and stained with haematoxylin and Cosin stain **Bancroft and Stevens, (1996).** Histological studies were monitored by microscopic examination of paraffin embedded slices of liver, kidney and brain from rats. All sections are examined at 200X magnification using a light microscope in Faculty of Veterinary, Cairo University, Egypt.

Statistical analysis

All data were analyzed by using the Statistical Package for Social Science (SPSS) version 17.00 to perform an ANOVA according to **SPSS, (2008).** The means of treatments were considered statistically significant at the 5% level ($P<0.05$), using Duncan test and the results were expressed as means ± standard deviation (SD) according to **Duncan, (1955).**

Results And Discussion

The chemical composition of molokhia leaves powder

Data in Table (2) displayed that the molokiha leaves powder had high contents of fiber, ash and protein. So, it had high nutritional value. The chemical composition of molokiha was in harmony with those of **Morsy *et al.*, (2015).** On the other hand, these results weren't in agreement with those of **Idirs *et al.*, (2010).** The results of difference in the chemical composition might be due to variation of kinds and seasons.

Table (2): The chemical composition of molokhia leaves powder on (dry weight basis)

Component	Moisture %	Dry matter	Protein %	Ash %	Fiber %	Oil %	Carbohy drate (Total Sugars) %
Molokhia leaves powder	9.1	90.9	24.99	14.2	45.72	2.19	12.9

Phenolic compounds and vitamins of molokhia leaves powder

Data presented in Table (2) showed that the molokhia leaves powder had high contents of total phenol, phenolic compounds and vitamins. The phenolic compounds contents were conformed to those of **Ademiluyi *et al.*, *(*2014).**

Table (3): Phenolics compounds and vitamins of molokhia leaves powder on (dry weight basis)

Phenolic compounds	Compounds (mg/100g)	vitamins	Compounds (mg/100g)
Total phenol	498	Nicotinic acid (Niacin)	898
Gallic	0.42	B6	204
Pyrogallol	23.30	Pyridoxin	137
4-Amino-benzoic	0.64	Folic acid	226
Protocatchuic	29.96	B12	496
Catechein	16.81	Vit.A	145
Chlorogenic	33.51	Vit.C	79
Catechol	7.69	Vit.D	13
Caffeine	3.56	Vit.E	0.42
P-OH-benzoic	7.01		
Caffeic	0.81		
Vanillic	3.37		
P-coumaric	4.56		
Ferulic	4.11		
Iso- Ferulic	0.85		
Ellagic	5.94		
Benzoic	15.15		
Coumarin	1.26		
3,4,5-methoxy-cinnamic	0.93		
Salycilic	8.03		
Cinnamic	0.55		

The results of vitamins contents of molokhia leaves powder were in accordance with those of **Yokoyama *et al.*, (2014)** also, the results of vitamin A, vitamin C, thiamine, niacin, riboflavin and nicotinamide contents of the molokiha leaves powder were in agreement with those of **Adeniyi *et al.*, (2012) and Ademiluyi *et al.*, (2014)**.

Biological assay

Feed intake, body weight gain and Feed efficiency ratio of lead poisoning rats feeding on molokhia leaves powder

Data in Table (4) showed the effect of feeding lead poisoning rats on 5%, 10 and 15% of molokhia leaves powder in feeding and growth parameters as feed intake (FI), body weight gain (BWG) and feed efficiency ratio (FER). The results indicated that the positive control group had the lowest values of these parameters compared to other groups. This was due to toxicity of lead that caused metabolism disturbance. These results were in line with those of **Okediran *et al.*, 2009, Ibrahim *et al.*, (2012)** and **Dewanjee *et al.*, (2013**) they reported that the Pb-acetate (5mg/kg body weight) for remedied rats shown a significant decreasing in weight. These results were due to inhibition of co-enzymes Q, antioxidant enzymes and decreased glutathione levels in the tissues. Moreover, the extent of lipid peroxidation, DNA fragmentation and haematological parameters were significantly changed in the Pb-acetate remedied rats as compared to control group.

Table (4) Feed intake, body weight gain and Feed efficiency ratio of lead poisoning rats feeding on molokhia leaves powder

Parameters / Groups	Feed intake (FI)	Body weight gain (BWG)	Feed efficiency ratio (FER)
Negative (-)	18.98±2.01a	45.06 ± 2.45a	2.27 ± 0.23a
Positive (+)	15.34±1.78c	24.68 ± 1.95c	1.52 ±0.12d
Molokhia 5%	16.36±1.9bc	25.91 ± 3.54c	1.61 ±0.74c
Molokhia 10%	16.6±0.93b	28.63 ± 2.40bc	1.66 ± 0.38bc
Molokhia 15%	17.4±1.08ba	30.9±1.22b	1.73±0.25b

Means in the same column with different superscript differ significantly at $p< 0.05$

Moreover, the group was fed on 15% molokhia leaves powder had the highest parameters comparing to other feeding groups. These results can be attributed to the fact that molokiha leaves had high levels of vitamins and minerals that plays a part in stimulating the rats appetite and can improve growth performance of rats when fed for eight weeks. These results were in conformity with those of **Ndlovu and Afolayan, (2008)**, **Wang *et al.*, (2011)** and **Kunle *et al.*, (2017)**.

Table (5): Relative organ weights of lead poisoning rats feeding on molokhia leaves powder

Parameters / Groups	Liver	Kidney	Brain
Negative (-)	2.14 ± 0.05 e	0.62 ± 0.03c	0.60 ± 0.04 c
Positive (+)	2.91 ± 0.09a	0.89 ± 0.14 a	0.74 ± 0.03a
Molokhia 5%	2.84 ± 0.20ab	0.86 ± 0.07 a	0.70 ± 0.03a
Molokhia 10%	2.74 ± 0.11bc	0.75 ± 0.03 b	0.68 ± 0.06 b
Molokhia 15%	2.67 ± 0.05c	0.68 ± 0.05bc	0.62b±0.1c

Means in the same column with different superscript differ significantly at $p < 0.05$

Relative organ weights of lead poisoning rats feeding on molokhia leaves powder

Data found in Table (5) showed the effect of feeding lead poisoning rats with 5, 10 and 15% molokhia leave powder on relative organ weights of liver, kidney and brain. The results displayed that the positive control group had the highest relative organs weights of liver, kidney and brain compared to other lead poisoning rats groups. These results were due to it had the highest lead poisoning that caused increases in weights of organs as a result to lead poisoning that caused defective metabolism in organs. These results were compatible with those of **Ibrahim *et al.*, (2012)** they found that the weights of liver, kidneys, heart and spleen were influenced by lead acetate intake from food. There was a significant increase in the organ weight after the period of lsoaexperimental.

The weights of lead poisoning rats were decreased as the levels of feeding were increased. These results might be due to molokhia leaves powder contained polyphenol components that used as protective

compounds against to lead poisoning effects. These results were in harmony with those of **Alkiyumi *et al.*, (2012)** they reported that the protective effect of the molokhia leave extract in chemical-induced liver damage might be due to its modification on the effects of detoxification enzymes and its antioxidant and free radical scavenger. Moreover, it underlines a scientific basis for the conventional use of molokhia leave for the treatment the liver disorders.

Blood Lead concentrations of lead poisoning rats feeding on molokhia leaves powder

The Lead level in whole blood is the best indicator mainly of recent exposure, although there can be mutable input to total blood lead concentration from past gatherings of lead in the body **(Lamphear *et al.*, 1999 and Barbosa *et al.*, 2005)**.

Data in Table (6) showed that there was increased blood lead concentration of rats after four weeks compared to after two weeks for poisoning. These results were due to rapid absorption and transportation by blood to different tissues. These results were in conformity with those of **Okediran *et al.*, (2009)** and **Okediran *et al.*, (2016)**. Moreover, the results indicated that there was a decrease in the proportion of blood lead concentration after six and eight weeks as a result of feeding on different levels of molokhia leaves powder. These results were due to molokhia had high contents of vitamins, phenolics and flavonoids that had against lead induced toxic manifestation in blood.

Table (6): Blood Lead concentrations of lead poisoning rats feeding on molokhia leaves powder

Parameters / Groups	µg/dl			
	Poisoning period		**Treatment period**	
	After 2 weeks	**After 4 weeks**	**After 6 weeks**	**After 8 weeks**
Negative (-)	7.52 ± 1.55 f	7.20 ± 1 f	7.29 ± 0.16f	7.52 ± 0.55f
Positive (+)	63.06 ± 1.55a	150.96 ± 1.01a	289.98 ± 0.47a	463.20 ±1.55a
Molokhia 5%	60.77 ± 1.55 a	145.00 ± 1.02 a	44.60 ± 1.04 b	32.28 ± 1.96 b
Molokhia 10%	61.80 ± 1.55a	147.60 ±0.9 a	43.09 ± 1.55 b	24.20 ± 1.55c
Molokhia 15%	61.80 ± 1.55 a	149.60 ±0.9 a	32.5 ±0.45d	19±0.98 d

Means in the same column with different superscript differ significantly at $p< 0.05$

On the other hand, these periods or quantity of feeding on molokhia leaves powder were not enough to reach safe levels on blood lead concentration. So, extract of molokiha leaves does have high effects of reducing blood lead concentration. These results were in harmony with those of **Dewanjee *et al.*, (2013) and Akinwumi *et al.*, (2016)** they reported that aqueous extract of molokiha leaves in lead clearance and could significantly restore the hematological parameters near to the normal status through antioxidant activity. This was due to extract of molokiha leaves is rich in flavonoids, saponins, anthraquinones, terpenoids, and phenols may be responsible for the observed protective role against lead intoxication. Suggest that has a high potential in the therapy/management of chromate-induced toxicities.

Liver functions of lead poisoning rats feeding on molokhia leaves powder

Liver enzymes (AST and ALT) are considered as an essential biomarker for the exposure of lead hepatotoxicity (**Haouas *et al.*, 2014).**

Data found in Table (7) displayed the serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) of lead poisoning rats that were fed on molokhia leaves powder. The results showed that the positive control group had the highest level of liver enzymes (AST and ALT). These results might be due to mainly to the leakage of these enzymes from the liver cytosol into the blood stream as reported by **Concepción *et al.*, (1993), Hassoun and Stohs (1995) and Haouas *et al.*, (2014)**

Table (7): Liver functions of lead poisoning rats feeding on molokhia leaves powder

Parameters / Groups	AST (IUI/I)	ALT (IUI/I)
Negative (-)	22.29 ± 0.95 e	25.57 ± 1.60 c
Positive (+)	88.43 ± 2.81 a	49.57 ± 1.78 a
Molokhia 5%	72.00 ± 1.48 ab	43.00 ± 2.62 a
Molokhia 10%	57.00 ± 1.03 c	32.00 ± 2.06 b

Means in the same column with different superscript differ significantly at $p < 0.05$

Also, AST and ALT enzymes were gradually decreased as affected by increasing levels of feeding on molokiha after eight weeks. So, the group feeding on 15% molokiha leave powder had the lowest levels of liver enzymes compared to other groups feeding on molokhia. On the other hand, the levels of liver enzymes did not reach to normal case. These results might be due to the rats sill had high levels of lead concentration as found in Table (6). These results of groups feeding on molokiha leave powder could be due to the antioxidant activities of molokiha leave that contain various amounts of phenol and flavonoids that caused gradually decreased in enzyme activities AST and ALT compared to intoxicated animals as found by **Iweala and Okedoyin (2014)** and **Adedosu *et al.*, (2015)**. Also, all these results are confirmed with those of **Omeje *et al.*, (2016) and Taiwo *et al.*, (2016)** they found that the molokiha leave extract was significantly (p=0.05) reduced the AST and ALT enzyme activities when compared to the control group at all doses tested and it had an influence on liver enzymes for protecting the liver cells and protection of liver from tumour.

Kidney functions of lead poisoned rats feeding on molokhia leaves powder

The metabolic wastes were removed by the kidney as, the concentration of which is usually required to assess the normal functioning of different parts of the nephrons **(Pari and Uma, 2000).**

Table 8: Kidney functions of lead poisoning rats feeding on molokhia leaves powder

Parameters / Groups	Urea nitrogen (mg/dl)	Creatinine (mg/dl)	Uric acid (mg/dl)
Negative (-)	55.57 ± 5.53 e	0.8 ± 0.11c	1.53 ± 0.20 d
Positive (+)	88.57 ± 1.51 a	1.63 ± 0.11a	2.94± 0.67 ab
Molokhia 5%	74.86 ± 5.52 b	1.60 ± 0.20 a	2.79 ± 0.38 b
Molokhia 10%	66.86 ± 5.70 c	1.51 ± 0.09 ab	2.73 ± 0.34 b
Molokhia 15%	62.14 ± 9.37 c	1.42 ± 0.18 b	2.07 ± 0.25 c

Means in the same column with different superscript differ significantly at $p < 0.05$

Data tabled in Table (8) showed that urea nitrogen, creatinine and uric acid were decreased as levels of feeding on molokiha leaves were increased compared to the positive control group. The results of the positive control group were due to the kidney functions is abnormal. So, creatinine increased as a result of decreased excretion of creatinine in the urine **(Nissl and Terra, 2004 and Hecht, 2006).** The results of group feeding on molokhia were might be due to improve in kidney functions. These might be due to molokhia had vitamins, phytochemical that repair the nephrons at the tubular and glomerular level that destroy as affected by lead poisoning. All these results were in line with those of **Bidaran, (2010)** and **Nikpey *et al*., 2013)** they found there are nutritional factors, such as some essential elements, vitamins, and antioxidants in preventing lead toxicity. Furthermore, the influence of competitive nutritional and antioxidant influence against lead cation inhibit the lead absorption and lead – induced oxidative stress in the body.

Histopathological examination

Histopathological examination of kidneys of lead poisoning rats feeding on molokhia (*C.olitorius*) leaves powder:

Kidneys of rats from group Molokhia 15% revealed protein cast in the lumen of some renal tubules (Fig.1). Other sections from group Molokhia 10% revealed congestion of intertubular blood vessels (Fig.2). However, kidneys of rats from group Molokhia 5% revealed protein cast in the lumen of renal tubules and vacuolation of endothelial lining glomerular tuft (Fig.3). While, kidneys of rats from positive group revealed periglomerular fibroblasts proliferation and inflammatory cells infiltration as well as thickening of the parietal layer of Bowman‟s capsule (Fig.4), granular degeneration of epithelial lining renal tubules, renal cast in the lumen of renal tubules (Fig. 5) . However, kidneys of rats from negative group revealed the normal histological structure of renal parenchyma (Figs. 6 and 7).

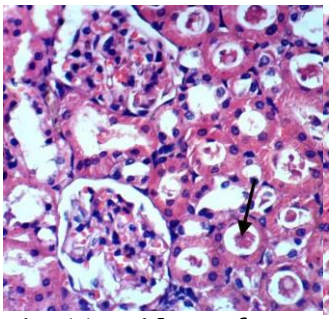

Fig. (1): Kidney of rat from group feeding on 15%Molokhia

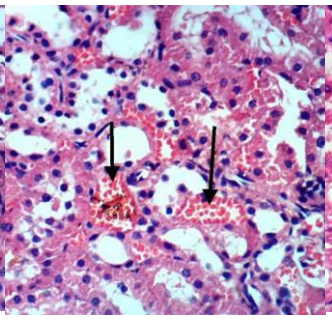

Fig. (2): Kidney of rat from group feeding on 10% Molokhia

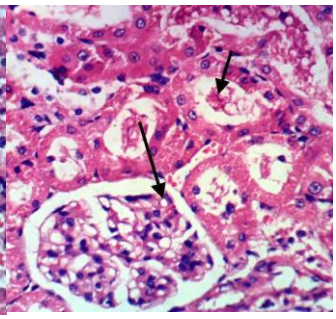

Fig. (3): Kidney of rat from group feeding on 5% molokiha

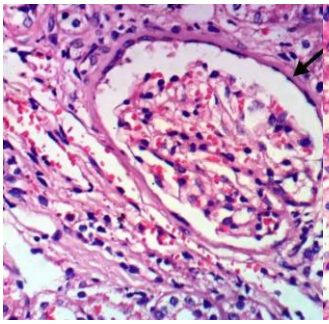

Fig. (4): Kidney of rat from positive control group

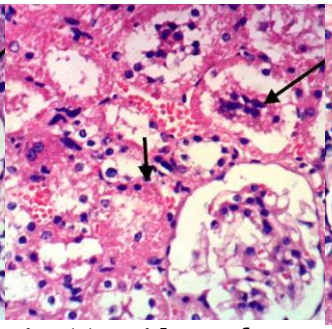

Fig. (5): Kidney of rat from group positive control group

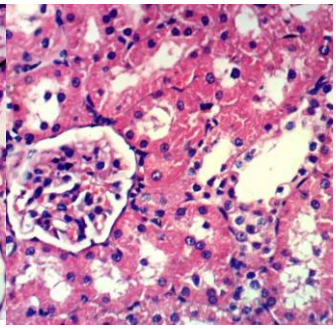

Fig. (6): Kidney of rat from negative control group

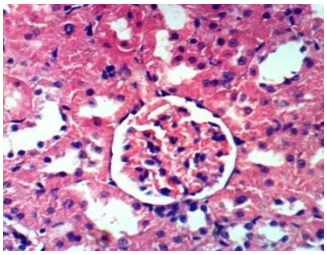

Fig. (7): Kidney of rat from negative control group

Histopathological examination of the liver of lead poisoning rats feeding on molokhia (*C.olitorius*) leaves powder

Lead Acetate poisoining caused hepatotoxic that increase liver enzymes compared to control group. The histological study indicated that lead can prompt several changes as hypertrophy of hepatocytes, lymphocytic infiltration, vacuolation, central vein dilatation and portal space. Tunel assay indicated significant DNA fragmentation in rats exposed to lead and demonstrated that Tunel assay can utilized to determinate DNA fragmentation in physical cells of rat liver **(Haouas *et al.*, 2014)**

Liver of rats from group Molokhia 15% revealed cytoplasmic vacuolation of hepatocytes (Fig.8) and focal hepatic necrosis associated with inflammatory cell infiltration (Fig.9). Liver of rats from group Molokhia 10% revealed kupffer cell activation (Figs.10 and 11). Liver of rat from group Molokhia 5% showing dilatation and congestion of hepatic sinusoids (Fig.12), focal hepatic necrosis associated with inflammatory cell infiltration (Fig.13) and congestion of hepatoportal blood vessel and few strands of fibroblasts in the portal triad (Fig.14). Liver of rat from group Positive showing cytoplasmic vacuolation of hepatocytes and fibroplasia in the portal triad (Fig.15) and focal hepatic necrosis associated with inflammatory cell infiltration (Fig.16). However, liver of rats from group negative revealed the normal histological structure of hepatic lobule (Figs.17, 18 & 19).

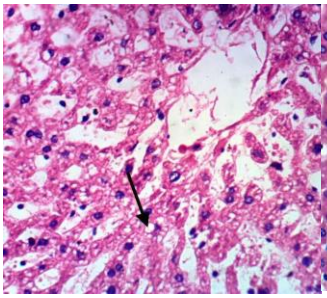

Fig. (8): Liver of rat from group feeding on 15%Molokhia

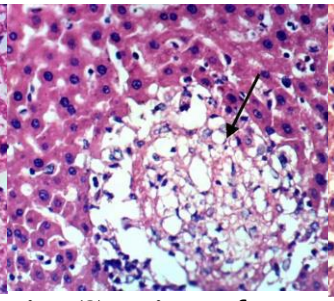

Fig. (9): Liver of rat from group feeding on 15%Molokhia

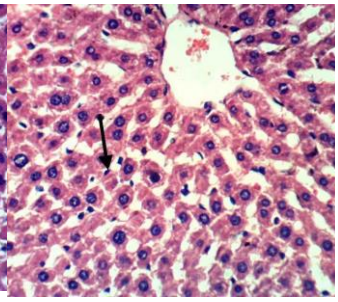

Fig. (10): Liver of rat from group feeding on 10%Molokhia

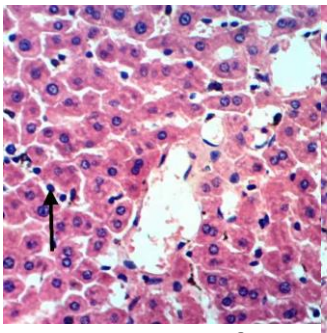

Fig. (11): Liver of rat from feeding on 10%Molokhia group

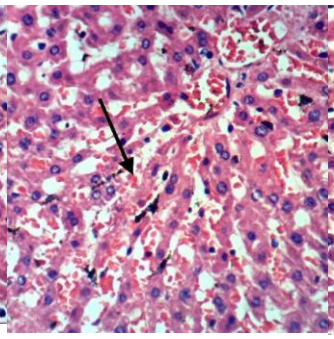

Fig. (12): Liver of rat from group feeding on 5%Molokhia

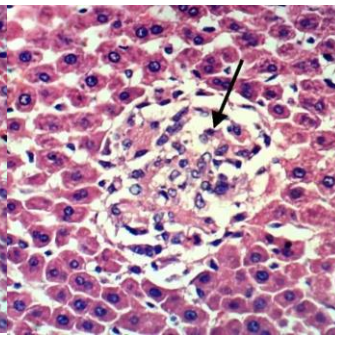

Fig. (13): Liver of rat from group feeding on 5%Molokhia

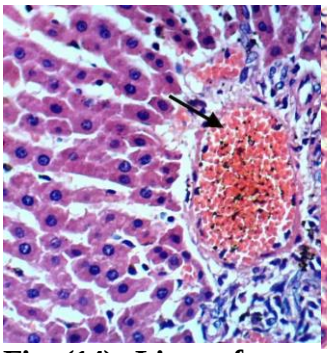

Fig. (14): Liver of rat from group feeding on 5%Molokhia

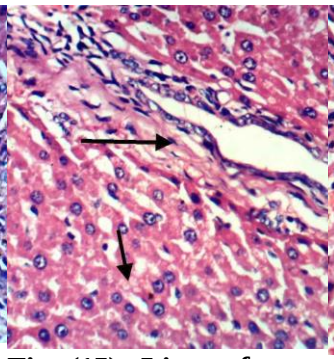

Fig. (15): Liver of rat from Positive group

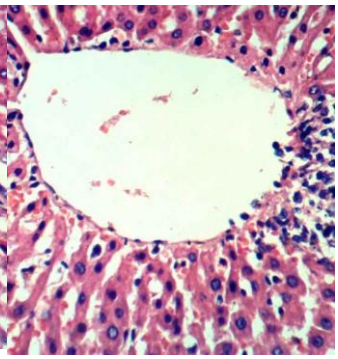

Fig. (16): Liver of rat from Positive group

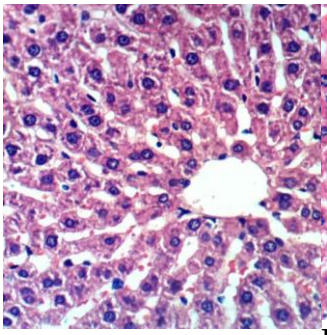

Fig. (16): Liver of rat from Positive group

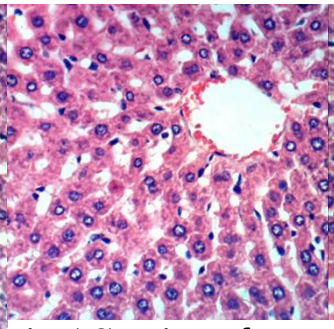

Fig. (18): Liver of rat from Negative group

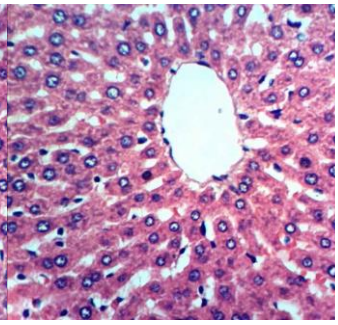

Fig. (19): Liver of rat from Negative group

Histopathological examination of the brain of lead poisoning rats feeding on molokhia (*C.olitorius*) leaves powder:

Brain of rats from group feed on Molokhia 15% necrosis of neurons and neuronophagia (Fig.20). Brain of rats from feed on Molokhia 10% showed necrosis of some neurons (Fig. 21). Meanwhile, brain of rats from group feed on Molokhia 5% revealed focal gliosis (Figs. 22). Moreover, brain of rats from positive group showed necrosis of neurons and multiple focal haemorrhage (Fig. 23). However, brain of rats from negative group revealed no histopathological changes (Figs. 24).

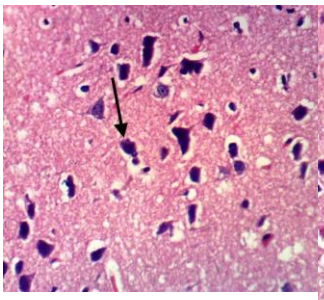

Fig. (20): Brain of rat from group feed on 15% Molokhia

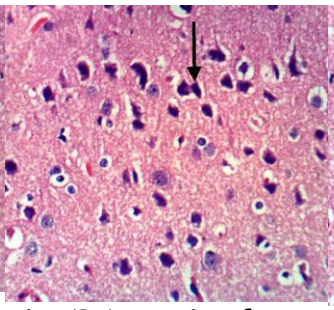

Fig. (21): Brain of rat from group feeding on 10% molokiha

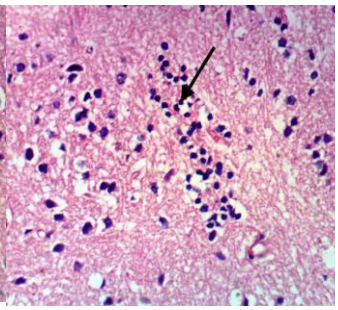

Fig. (22): Brain of rat from group feeding on 5% molokiha

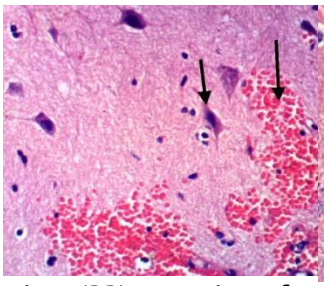

Fig. (23): Brain of rat from positive group

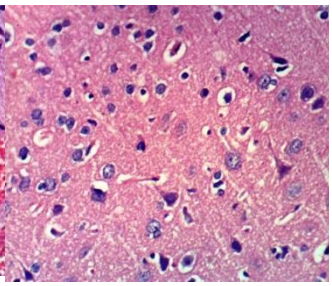

Fig. (24): Brain of rat from negative group

Conclusion

Molokhia (*C.olitorius*) leaves powder has high nutritional value. It had high contents of fiber, vitamins and total phenol compounds that

had high antioxidant activity and antitumor of organ tissues of kidney, liver and brain. So, it can be used as an anti-poisoning for Lead poisoning.

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تأثير التغذية بمسحوق أوراق الملوخية على الفئران المصابة بتسمم الرصاص

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الملخص

إن التسمم بالمعادن الثقيلة وخاصة تسمم الرصاص له تأثيرات ضارة على صحة الإنسان. و بالإضافة هناك العديد من الأبحاث التى أثبتت ان لبعض النباتات تأثيرات مضادة لتسمم المعادن. لذلك تم هذه البحث لدراسة مدى تأثير تغذية الفئران المصابة بتسمم الرصاص على ٥, ١٠, ١٥% مسحوق أوراق الملوخية لمدة ثمانية أسابيع على تركيز الرصاص فى الدم و على وظائف الكبد والكلى. ودراسة الفحص الهستولوجى لأنسجة الكى والكبد والمخ. أظهرت النتائج إرتفاع محتوى مسحوق أوراق الملوخية من البروتين, الألياف, الفينولات الكلية و الفيتامينات. بالإضافة الى ذلك و نتيجة لتغذية الفئران المصابة بتسمم الرصاص على مسحوق أوراق الملوخية, أظهرت المجموعة المغذاة على ١٥% مسحوق أوراق الملوخية أعلى مستويات من معدل المنتاول من الغذاء, و المكتسب من الوزن, و معدل الإستفادة من الغذاء بالمقارنة بالمجموعة الموجبة لسمية الرصاص و المجموعات المغذاة الأخرى. كما أوضحت النتائج حدوث إنخفاض تدريجي فى تركيز الرصاص فى الدم, وإنزيمات الكبد, و وظائف الكلى (نيتروجين اليوريا, و الكرياتينين وحمض اليوريك) فى الفئران المصابة بتسمم الرصاص وذلك بزيادة مستويات التغذية على مسحوق أوراق الملوخية. ولذلك المجموعة المغذاة على ١٥% مسحوق أوراق الملوخية لديها أقل مستويات من هذه التحاليل بالمقارنة بالمجموعة الايجابية والمجموعات الأخرى المغذاة على الملوخية. كما أظهرت نتائج الفحص الهستولوجى لأنسجة الكلى و الكبد والمخ أن المجموعة المغذاة على ١٥% مسحوق أوراق الملوخية كانت أقل المجموعات تعرضا لخطر التسمم بالرصاص بالمقارنة بالمجموعة الايجابية والمجموعات الأخرى المغذاة على الملوخية ولكنه ليس كاف لمعالجة كل التغيرات التى حدثت فى أنسجة هذه الأعضاء نتيجة للتسمم بالرصاص. أخيرا، مسحوق أوراق الملوخية تحتوى على كمية كبيرة من الألياف, و الفينولات الكلية و الفيتامينات التى لها نشاط عالى مضاد للاكسدة و التورم لأنسجة الكلى, و الكبد, والمخ. ولذلك يمكن أن تستخدم كمضاد للتسمم الرصاص.

الكلمات الدالة: ملوخية ، سمية, الرصاص ، الفيتامينات, الفينولات الكلية, ، الكبد. الكلى, المخ ، الفحص النسيجي