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Abstract

The purpose of this study was to investigate the nutritional effects of milk thistle extract and its efficacy against lead toxicity in rats. The experiment used 30 albino rats weighing 140±5g, divided into five groups (6 rats per group). The first group was a negative control, and the second group was a positive control fed a basal diet supplemented with 200 mg/kg lead acetate. Groups (3 to 5) were fed the same lead-supplemented diet with an additional 100 mg, 300 mg, and 500 mg/kg of milk thistle extract, respectively, for six weeks. The chemical composition of milk thistle, weight gain, and levels of calcium, phosphorus, lead, kidney function, liver enzymes, malondialdehyde and some oxidative enzymes were measured.

The results showed an increase in weight in the rats treated with milk thistle extract compared to the positive control group. Liver enzymes and kidney functions significantly decreased in groups (3 to 5) compared to the positive control group. There was a significant improvement in serum calcium and phosphorus levels in the groups treated with milk thistle extract. Lead levels decreased in all three concentrations of milk thistle extract used compared to the positive control group. Groups (3 to 5) showed an increase in catalase levels and a decrease in malondialdehyde due to the consumption of milk thistle extract. Therefore, this study recommends that the consumption of milk thistle extract at the three different levels has a positive effect on health by reducing the harmful effects of lead.

Key Words: Lipid profile, Lead toxicity, Liver enzymes, Milk thistle extract.

Introduction

The persistent and continuous presence of environmental pollutants, including lead, which is a hazardous heavy metal, continues to pose a significant burden & difficult task in numerous developing nations **(Sunday *et al*., 2012).** Living organisms' cells had division to substitute old cells, develop to become specialized, & had apoptosis in response to harm. These processes are rigorously controlled to guarantee the continuous existence of life and a robust system. Nevertheless, if there is an imbalance or disruption in any of these processes, it can lead to various illnesses. Over time, environmental pollution has persistently caused several health problems worldwide, resulting in an increased economic burden on countries internationally **(Landrigan and Fuller, 2015) .**

Lead (Pb) is a prevalent environmental pollutant that contributes to the development of several human diseases by reducing defense mechanisms of antioxidant & causing genotoxic impacts **(Gagan *et al*., 2012).** A medical humanitarian organization has reported that the unlawful mining of gold ore in the northern portion of Nigeria (specifically Zamfara State) has resulted in a dangerous level of lead poisoning in the soil and household dust. According to **Medecins San Frontieres (2012)**, this resulted in a mortality rate of up to 40% among children and caused the death of over four hundred lives .

Lead is a pervasive environmental contaminant that has been documented to cause pathological disorders in people **(Victor *et al*., 2020).** Oxidative stress is a potential method via which lead toxicity can occur. The exposure to lead in living organisms enhances oxidative stress induction through 2 simultaneous pathways. The first pathway involves the generation of reactive oxygen species, for example singlet oxygen, hydroperoxides, & hydrogen peroxide. The second pathway involves the lessening of antioxidant reserves by the generated reactive oxygen species **(Patra *et al*., 2011& Lopes *et al*., 2016).** Oxidative stress can be characterized by the failure to promptly remove the reactive intermediates produced or to effectively repair the resultant

damage **(Flora, 2009 & Elias *et al*., 2014).** The negative consequences resulting from the utilize of artificial medications prompted the exploration of ethno medicinal plants that possess the ability to effectively treat lead toxicity .

Silybum marianum, often known as milk thistle or MT, is a plant belonging to the asteraceae family. It has a long history of usage as a therapeutic plant, dating back to ancient times **(Siegel and Stebbing, 2013).** Silymarin, composed of distinct antioxidant flavonolignans, is the primary bioactive constituent of milk thistle. Silymarin is recommended for the prevention of liver, biliary tract problems, oncological conditions, as supportive therapies for hepatitis C, HIV, diabetes, and hypercholesterolemia. It is also known to enhance lactation **(MacDonald-Ramos *et al*., 2020& Javeed *et al*., 2022).** MT exhibits considerable potential for many uses, including but not limited to human consumption & industrial utilization. These applications encompass the use of MT in dermatological & cosmetic formulations, as highlighted by (**Marceddu *et al*., 2022 & Kim *et al*., 2023).** Achenes, which are more frequently referred to as seeds, are the parts of the plant that take the highest concentration of silymarin. On the other hand, some components of the plant have also been utilized as traditional remedies **(Chambers *et al*., 2017).** The milk thistle, which was initially discovered in Southern Europe, Asia Minor, the Southern Russian Federation and Northern Africa, is now extensively dispersed over the globe in locations that are both warm & dry. Europe, Asia, North & South America, & Southern Australia are the primary regions where it is cultivated for the purpose of producing silymarin. **(Drouet *et al*., 2018).**

Silybum marianum, which is sometimes referred to as milk thistle, is an herb that is in high demand and is used as a supportive therapy for liver disorders. According to **Raclariu-Manolica and Socaciu, (2023)** the principal biologically active ingredient that can be discovered in MT is silymarin, which is composed of several flavonolignan metabolites. The principal bioactive component of milk thistle (MT) is silymarin, which is a combination of seven flavonolignan isomers. These flavonolignan isomers are silybinin A & B, isosilybinin A & B, silychristin A &

B and silydianin. These isomers can be discovered in distinct parts of the MT plant, involving roots, leaves, fruits, & predominantly in the seeds **(Javeed *et al*., 2022 & Aziz *et al*., 2021).** Furthermore, the silymarin complex also includes a polyphenolic component that has the potential to be bioactive. This chemical is commonly known as a "polymeric fraction" and makes up 30% of the complex **(Biedermann *et al*., 2014).** MT also contains various flavonoids such as apigenin, taxifolin, naringin, luteolin, eriodyctiol, chrysoeriol, kaempferol, quercetin, rutin and dihydrokaempferol. It also contains fatty acids like oleic, linoleic, behenic, palmitic acids and sterols such as cholesterol, campesterol, stigmasterol, and sitosterol. Additionally, MT contains proteins and sugars including rhamnose, arabinose, xylose, and glucose (**Abenavoli *et al*., 2018).** Silybin, which makes up fifty to seventy percent of the silymarin extract, is the most biologically active component and has been extensively researched as the primary flavonolignan **(Loguercio, 2011).** silybin B, Silybin A, & isosilybin A were found in cultures of Aspergillus iizukae, a type of fungus that grows inside the leaves of MT **(El-Elimat *et al*., 2014).**

The silymarin complex consists of seven primary flavonolignans & the flavonoid taxifolin. These compounds are currently being utilised as markers for the authentication & quantification of MT. Their testing is conducted according to the guidelines provided in specific monographs, such as those outlined in the European & United States Pharmacopoeias, among others **(Aziz *et al*., 2021& Bijak, 2017).**

Aim of the study

This study was conducted to evaluate the importance of a milk thistle diet and its effectiveness in combating lead toxicity in rats.

Materials and Methods

Materials:

1-Chemicals:

Vitamins, casein, cellulose, minerals, starch and choline have been obtained from the Morgan Chemical Factory in Cairo,

Egypt. Lead acetate was purchased from Sigma Chemical Company, Cairo, Egypt.

2-Plant:

Milk thistle was obtained from Agriculture Research Center, Egypt.

3-Rats:

Thirty albino rats of the Sprague-Dawley strain, each weighing approximately 140 ± 5 grams, were donated by the Agricultural Research Centre of the Health Research Institute in Giza, Egypt.

4-Kits:

The kits used for biochemical analysis were sourced from the Biodiagnostic Company for Pharmaceutical and Chemicals in Dokki, Egypt.

Methods:

1-At the Agriculture Research Centre, an identification process was performed to determine the specific species of *Silybum marianum L.* within the plant kingdom plant.

Rank	Scientific Name
Kingdom	*Plantae*
Division	*Tracheophytes*
Subdivision	*Angiosperms*
Class	*Eudicots*
Superorder	*Asterids*
Order	*Asterales*
Family	*Asteraceae*
Subfamily	*Carduoideae*
Genus	*Silybum*
Species	*Silybum marianum*

2-Chemical analysis of milk thistle:

The milk thistle dried samples underwent chemical analysis to determine their protein, fat, moisture and ash levels. The analysis was conducted using the procedures outlined by **A.O.A.C. (2005).** The crude fiber contents were assessed using enzymatic and gravimetric methods as specified by the **A.A.C.C. (2000).** The

total carbs have been found utilizing the difference method, as stated by (**Pellet and Sossy, 1970)**

3-Preparation of milk thistle extract:

The desiccated seeds of MT were ground into a fine powder, and five grams of the powder were extracted with ninety-five percent methanol (100 ml) three times, with continuous stirring, at twenty-eight degrees overnight. The combined extracts were evaporated to a known volume under decreased pressure. The residual solvent was evaporated to dryness on a water immersion at 40 degrees Celsius **(Aslam *et al*., 2012).**

4-Ethics approval:

The experimental procedure for this research was permitted by the Institutional Animal Care & Utilization Committee (ARC-IACUC) at the Agricultural Research Center in Egypt (approval number: ARC/AHRI/88/23).

5-Preparation of basal diet:

Reeves *et al*., (1993) were employed to formulate the basal diet. The product is composed of twenty percent protein, ten percent sucrose, two percent choline chloride, 4.7 percent corn oil, 3.5 percent minerals mixture, one percent vitamin mixture, & five percent fiber. Corn starch comprised the remaining part, which was increased to one hundred percent.

6-Experimental procedure:

Following the period of adaptation, 30 male albino rats of the Sprague-Dawley strain, weighing roughly 140 ± 5 grams, were divided randomly into five main groups, each with six rats. The first group was fed on basal diet as(–ve). The remaining twenty-four animals were subjected to lead toxicity (utilizing lead acetate) at a rate of two hundred mg/kg in accordance with **Newairy and Abdou, (2009)**. The second group served as the positive control . Groups three to five were fed varying concentrations of milk thistle extract. The following was the order in which all divisions were assigned:

Group one: Rats were maintained on a basal diet throughout the experimental duration as(–ve).

Group two: Rats were exposed to lead toxicity and fed on basal diet as (+ve).

Group three: Rats were maintained on a basal diet addition to 100 mg / kg of body weight milk thistle extract in the mouth.

Group four: Rats were maintained on a basal diet supplemented with 300 mg / kg body weight milk thistle extract in the mouth.

Group five: Rats were maintained on a basal diet supplemented with 500 mg / kg body weight milk thistle extract in the mouth.

The animals were confined in the Animal House of the Agricultural Research Centre, Giza, Egypt, and were maintained in hygienic conditions at a room temperature of 25 ± 2 degrees celsius and a moderate humidity of fifty to sixty percent. Water and basic sustenance were permitted on an ad libitum basis.

7-Blood samples:

The abdominal aorta was used to obtain blood samples following twelve hours of fasting at the conclusion of the experiment (six weeks), during which the rats were scarified under ether anesthesia. Clean, sterile centrifuge tubes were used to receive blood samples. The tubes were then allowed to clot at room temperature before being centrifuged at three thousand revolutions per minute for ten minutes to separate the serum. The serum was aspirated carefully, transferred to sterile cuvette tubes and frozen at minus twenty degrees Celsius. for analysis. The subsequent parameters were determined by analyzing all serum samples.

Identification of Serum Liver Enzymes: The alanine aminotransferase (ALT) enzyme was quantified in serum using the method of **Sherwin (1984). Young (1990)** was employed to identify the enzyme aspartate aminotransferase(AST).

Identification of Serum Alkaline Phosphatase Concentration: The measurement of serum alkaline phosphatase (ALP) was conducted using the procedure defined by **Roy (1970).**

Identification of Serum Kidney Functions: The concentration of serum urea nitrogen was measured using the **Fossati *et al*., (1980)** technique. The determination of creatinine was conducted via the method published by **Henry (1974)**, whereas the measurement of uric acid was performed following the method developed by **Schultz (1984).**

Identification of Serum Lead: The method of **(Parsons, 2001)** was used for determining the serum lead concentration.

Identification of Serum Malondialdehyde (MDA): The determination of MDA was conducted following the method described by **Draper and Hadly, (1990)**.

Identification of Serum Catalase (CAT): The activity of CAT was determined in tissue homogenate using the method described by **Aebi, (1984).**

Identification of Serum Calcium and Phosphorus:

Aloia *et al*., (1984) method was employed to determine the concentration of calcium and phosphorus in serum.

Identification of Serum Lipid Profile:

Fossati and Prencie (1982) methodology was used to assess the serum triglycerides (TG). Serum total cholesterol (TC) was measured using **Henry *et al*., (1974)** technique. According to **Burstein, (1970)** the amount of high density lipoprotein cholesterol (HDL-C) in the serum was measured. **Friedewald *et al*., (1972)** methodology was used to determine the concentrations of low density lipoprotein cholesterol (LDL-c) and Very low-density lipoprotein cholesterol (VLDL-c) in through the following computations:

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

VLDL-c (mg/dl) = (Triglycerides /5)

8-Statistical analysis:

The data were provided as the mean value ± the standard error. The statistical analysis was performed utilizing SPSS, a computer-based statistical software (Version 18.0 SPSS Inc., Chicago, USA), and the Dunk 'test multiple range post-hoc test was employed. The data was examined using an ANOVA. The data were determined to be statistically significant at a significance level of P <0.05 as published by (**Snedecor and Cochran, 1980).**

Results and Discussion

Table (1): Composition of dehydrated milk thistle.

Chemical composition (%)	Milk Thistle
Protein	30.09
Fat	19.74
Carbohydrates	6.8
Moistures	6.27
Ash	2.37
Fiber	7.4

The information presented in Table (1) clearly indicated that the dried milk thistle had greater concentrations of protein, fat ,carbohydrates, moisture, ash and crude fiber contents. *Silymarin*, the lipophilic extract derived from the seeds of MT , is the active complex **(Abenavoli *et al.*, 2010).** *Silymarin* is composed of several flavonolignans, which are isomers of flavonolignans, as well as flavonoids and other components **(Albassam *et al*., 2017).** The seven main flavonolignans that can be employed as marker compounds for quantification assays of silymarin are silybins A & B, isosilybins A & B, silychristin A, isosilychristin and silydianin. Additionally, the flavonoid taxifolin can also be utilized for this purpose **(Abouzid *et al*., 2016).** Flavonolignans exhibit a wide range of structural variability due to the connection of the C6C3 unit to the flavonoid nucleus through C-C or C-O linkages at various places. This results in the formation of dioxane, cyclohexane rings, furan, or simple side chains on either side. Typically, these molecules have many chiral centers, therefore they commonly exist as stereoisomers in nature **(Soliman *et al*., 2018).** Flavonolignans are formed through the process of

oxidative coupling between a flavonoid called phenylpropanoid & taxifolin, typically coniferyl alcohol. This is followed by the coupling of the 2 radicals. Silybin is the predominant and most potent component among the isomers, comprising approximately sixty to seventy percent of the total. It is followed by silychristin at twenty percent, silydianin at ten percent and isosilybin at five percent **(Saller *et al.*, 2001).** In addition to, milk thistle also contains various other flavonoids (for example, dihydrokaempferol, quercetin, apigenin, kaempferol, eriodyctiol, naringin, chrysoeriol), proteins (twenty-five - thirty percent), sugars (rhamnose, arabinose, xylose, glucose), sterols, tocopherols (stigmasterol, campesterol, sitosterol, cholesterol) and lipids (15– 30 percent) in the form of triglycerides sixty percent linoleic, thirty percent oleic, & nine percent palmitic acid **(Abenavoli *et al.*, 2010).** Although *S. marianum* fruit lipids have a high nutritional value, the oil is regarded as an undesirable by product of *silymarin* production and must be eliminated from the fruits before *silymarin* extraction **(Abouzid *et al.*, 2017).**

Table (2): The impact of milk thistle extract on body weight gain, feed intake and feed efficiency ratio in lead intoxicated rats.

Parameters / Groups	BWG (%)	FER	Feed Intake (g/day)
Group (1) negative control	146.20±0.58^{b}	11.07±0.16^{a}	13.22±0.19^{a}
Group (2) positive control	132.20±1.02^{e}	10.15±0.23^{b}	10.04±0.28^{c}
Group (3) 100 mg/kg milk thistle extract	137.00±0.70^{d}	10.79±0.18^{b}	12.70±0.18^{b}
Group (4) 300 mg/kg milk thistle extract	148.80±0.58^{a}	11.25±0.21^{a}	13.24±0.24^{a}
Group (5) 500 mg/kg milk thistle extract	140.80±0.97^{c}	10.98±0.10$^{a, b}$	12.82±0.11^{b}

*Values are expressed as means ±SE.

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

The information presented in Table 2 indicates that the (+ve) group had a significant decline in body weight gain compared to the negative control group. Furthermore, the rats in groups three, four, and five exhibited a significant rise in body weight gain compared to the rats in the (+ve) group. The correlation between the rise in body weight and the elevation of milk thistle extract levels may be attributed to the rat's response to the odor , taste, and overall appeal of the food .

The body weight of rats exposed to lead when impaired was considerably lower compared to the body weight of the healthy

control group. The data clearly demonstrated that lead exposure resulted in a significant decline in b.w. g . The detrimental impact of lead on body weight growth increased in correlation with the rise of lead acetate dosages. There wasn't significant variation in the amount of food consumed by the four groups. This indicates that there wasn't a correlation between the amount of body weight gain, feed intake and feed efficiency ratio. Furthermore, the presence of lead acetate resulted in a decline in feed efficiency compared to the control group. This drop was observed alongside a rise in body weight gain, but not in feed intake. The present study found that the adverse impact of lead acetate consumption wasn't significantly enhanced as the amount of lead acetate increased.

The impact of lead acetate on feed intake, body weight gain and feed efficiency ratio showed a gradual rise over the whole investigation duration across all four distinct groups. The intoxicated rat with lead had a considerably lower ultimate b.w.g compared to the healthy normal group. The information clearly demonstrated that lead exposure resulted in a significant decline in b.w.g . The negative impact of lead on b.w g was heightened in correlation with the escalation of lead acetate dosages **(Ibrahim *et al.*, 2012) .**

The acquired results agree with the results of a prior investigation. The research performed by **Seddik *et al.*, (2010)** revealed that the intake of lead resulted in a decline in the growth rate of rats. The increase in body weight resulting from toxic ions can be attributed to various mechanisms, one of which is the disruption of zinc-dependent enzymes that play a crucial role in multiple metabolic processes, leading to an imbalance in metabolism **(Hwang and Wang, 2001).**

The findings from a prolonged investigation into the consumption of milk thistle seed oil (MTSO) indicate that there weren’t significant alterations in the ultimate body weights of rats fed two different amounts of MTSO in comparison to the control diet groups. The findings align with several animal experiments conducted on various seed oils, including those by **Moon *et al.*, (2001);Gorinstein *et al.*, (2003); Cintra *et al.*, (2006); Makni *et***

***al.*, (2009)&Visavadiya and Narasimacharya (2008).**

Table (3): **The impact of milk thistle extract on Liver and Kidney Relative weight in lead intoxicated rats.**

Parameters / Groups	Liver	Kidney
	%	
Group (1) negative control	2.88±0.17[b]	0.60±0.10[b]
Group (2) positive control	3.97±0.22[a]	1.11±0.08[a]
Group (3) 100 mg/kg milk thistle extract	2.91±0.12[b]	0.74±0.03[b]
Group (4) 300 mg/kg milk thistle extract	2.91±0.08[b]	0.57±0.10[b]
Group (5) 500 mg/kg milk thistle extract	2.72±0.15[b]	0.41±0.03[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 the results of modifications to the relative organ weight. Compared with the (-ve) group, rats in the positive group with lead toxicity exhibited a rise in the average value of the relative weights of the kidneys and liver. The (–ve) group exhibits a decline in the relative weight of the kidney and liver compared to the (+ve) group. In comparison to the positive control group, the average relative weight of the liver and kidney decreased in the group of positive controls which were supplemented with milk thistle extract at three levels.

The liver and kidney were less affected by lead. Parallel to the rising dose, these observations of Pb2+ ingestion are significantly enhanced. The observed increase in the weight or ratio of the organs was believed to be the result of necrosis and apoptosis, which could be attributed to the accumulation of lipids in the four organs. Pb2+ therapies resulted in a substantial accumulation of lipids in the kidney cells of rat **(Hwang and Wang, 2001).**

Table (4): **The impact of milk thistle extract on serum lipid profile in lead intoxicated rats.**

Parameters / Groups	Total cholesterol	Triglycerides	High-density lipoprotein cholesterol	low-density lipoprotein cholesterol.	Very low-density lipoprotein cholesterol
	(mg /dl)				
Group (1) negative control	104.44±1.18[d]	76.40±0.74[d]	54.60±0.74[a]	34.56±1.28[c]	15.28±0.15[c,d]
Group (2) positive	136.90±1.10[a]	142.80±1.39[a]	41.60±0.92[c]	66.74±0.42[a]	28.56±0.27[a]

control					
Group (3) 100 mg/kg milk thistle extract	122.18±0.83^{b}	122.80±0.86^{b}	51.80±0.66^{b}	45.82±1.47^{b}	24.56±0.17^{b}
Group (4) 300 mg/kg milk thistle extract	116.84±0.73^{c}	97.20±1.02^{c}	51.20±0.49^{b}	46.20±1.10^{b}	19.44±0.20^{c}
Group (5) 500 mg/kg milk thistle extract	111.34±0.80^{c,d}	91.40±0.74^{c,d}	53.20±0.58^{a}	38.86±2.13^{c}	18.28±0.15^{c}

*Values are expressed as means ±SE.

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

Table (4) illustrates the mean value of total cholesterol in rats that were administered a variety of diets. It was evident that the average of total cholesterol in the positive was 136.90±2.47(mg/dl), which was greater than that in the negative group (104.44±2.65(mg/dl). This difference was particularly pronounced when milk thistle was included in the rats. Specifically, group 5 (500 mg of milk thistle) exhibited the lowest TC level .

For TG, it was noted that the average of the positive control group was 142.80±3.11 (mg/dl), which was greater than negative control group 76.40±1.67 (mg/dl). In comparison to the (+ve) group, all treated groups with MT exhibited significant reductions in TG levels. The most favorable outcome was observed in group 5, which consisted of rats with lead toxicity who were administered five hundred milligrams of milk thistle .

The positive group had a mean HDL-c value of 41.60±2.07 (mg/dl), which was less than the negative group's mean of 54.60±1.67 (mg/dl). It was evident that the mean of HDL-c in the (+ve) group was 30±2.81, which was significantly less than that of the control (-ve) group. Nevertheless, when MT was incorporated into the rats' diet, a significant rise was observed in comparison to the positive group, particularly in the 5 group (500 mg of MT), which exhibited the highest level of high-density lipoprotein cholesterol .

Low-density lipoproteins-cholesterol the mean value of the (+ve) group was 66.74±0.94 (mg/dl) greater than that of the control (-ve) group, which was 34.56±2.88 (mg/dl). All the treated groups by milk thistle exhibited a significant reduction in LDL

levels in comparison to the (+ve) group.

Concerning VLDL-C The mean value of VLDL-c in the positive group was 28.56±0.62 (mg/dl), while in the negative group it was 15.28±0.33 (mg/dl). The distinction was significant when milk thistle was included in the rats diets, as a significant decline was detected in comparison to the positive group. This research is consistent with the findings of **(Dabbour *et al*., 2014) & (Amin *et al*., 2019)** who stated that the lipid profile was primarily influenced by the ratio of milk thistle seed oil. The administration of silymarin in rats resulted in a decline in the plasma concentrations of LDL, VLDL, TC and TG when compared to both hyperlipidemic rats and the negative group.

Silymarin effectively preserved high-density lipoprotein cholesterol at an elevated level and reduced low-density lipoprotein cholesterol to a lower level, resulting in notable improvements to the circulatory system. A critical protective function of silymarin in the mRNA regulation of genes involved with oxidative stress and lipid metabolism has been demonstrated through real-time PCR analysis **(Ni and Wang, 2016).** The current findings indicate that silybin is advantageous for hypertriglyceridemia, hyperglycemia supportive treatment and it significantly raised the protective HDL content. Nevertheless, the total cholesterol level was not impacted by its administration. **Porupa *et al*., (2015)** discovered that the micronized form of silybin resulted in a greater rise in protective High-density lipoprotein cholesterol levels.

Table (5): **The impact of milk thistle extract on serum concentrations of liver enzymes on lead intoxicated rats.**

Parameters / Groups	ALT	AST	ALP
	(U/L)		
Group (1) negative control	26.80±0.37^{e}	32.80±0.37^{e}	621.20±1.06^{d,e}
Group (2) positive control	66.40±0.74^{a}	85.00±0.89^{a}	852.60±1.43^{a}
Group (3) 100 mg/kg milk thistle extract	53.40±0.92^{b}	60.60±0.51^{b}	764.40±2.15^{b}
Group (4) 300 mg/kg milk thistle extract	42.60±1.03^{c}	54.80±1.24^{c}	715.80±1.77^{c}
Group (5) 500 mg/kg milk thistle extract	34.40±0.87^{d}	45.60±0.92^{d}	656.60±1.43^{d}

*Values are expressed as means ±SE.

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

Table (5) revealed the impact of MT leaves on the serum activity of alanine aminotransferase (ALT). In the positive group, the activity of alanine aminotransferase was markedly elevated in rats exposed to lead poisoning, with an average of 66.40±1.67 (U/L), as compared to the negative control group 26.80±0.83 (U/L), as indicated by the data. Nevertheless, the serum level activities of alanine aminotransferase were reduced in rats that were fed a MT-containing diet at three intake levels, as compared to the positive control group. The average of the serum level activities was 53.40±2.07 (U/L), 42.60±2.30 (U/L) and 34.40±1.94 (U/L), respectively. At group five, rats were treated with milk thistle, and their serum ALT activity was lowest, with an average of 34.40±1.94 (U/L).

Additionally, Table (5) illustrates the impact of milk thistle leaves on the activity of AST. Aspartate aminotransferase levels in serum were significantly elevated in the positive control group, which was exposed to lead toxicity, with a mean value of 85.00±2.00 (U/L) , in contrast to the negative control group, which had a mean value of 32.80±0.83(U/L).The results indicated that the serum activity of aspartate aminotransferase was reduced in rat that were fed milk thistle in the diet, regardless of the intake level, in comparison to the positive control group. The serum activity of AST was decreased in rat that were fed milk thistle in group five, with a mean value of 45.60±2.07 (U/L) .

Furthermore, Table (5) illustrates the impact of milk thistle on alkaline phosphatase (ALP). In comparison to the negative control group 621.20±2.38(U/L),the concentration of alkaline phosphatase in serum was significantly elevated when rat were exposed the toxicity of lead, with an average of 852.60±3.20 (U/L). The results showed that the mean value of alkaline phosphatase in serum was significantly reduced in the group of rat fed on milk thistle at three concentrations in the diet. The average was 764.40±4.82 (U/L),715.80±3.96 (U/L) and 656.60±3.20 (U/L), respectively, when compared to the positive group 852.60±3.20 (U/L) .

Our findings are consistent with **Kumar and Khanna (2018) and Amin *et al*. (2019)**, who found that all groups that were

administered carbon tetrachloride or glycerol/saline solution and treated with varying concentrations of dried MT (twenty percent to forty percent) had a significant decrease in liver function in comparison to the +ve group. In one study, *silymarin* premedication has been shown to prevent the hepatotoxic effects of acetaminophen, thallium tetrachloride and halothane **(Ross, 2008)**. In rats, *silymarin* inhibits liver enzymes for example ALT, GGT and AST **(Detaille *et al*., 2008).** In one research, *Silymarin* was able to decrease mortality rates in cases of alcoholic cirrhosis after four years. Conversely, *silymarin* was unable to decrease hepatic mortality in cirrhotic patients in a separate study **(Tsai *et al*., 2008).** Our research demonstrated that MT extract protects the liver from the adverse effects of lead toxicity .

Vargas-Mendoza *et al*., (2014) have identified that *silymarin* has a variety of beneficial impacts, particularly in the context of liver illnesses, due to its well-documented hepatoprotective properties. In experimental human hepatocytes, *silymarin* demonstrated beneficial effects on nonalcoholic steatohepatitis, nonalcoholic fatty liver illness and fibrosis. Silybin inhibits insulin resistance, oxidative stress and the accumulation of fat in the liver **(Federico *et al*., 2017; Marin *et al*., 2017).** It enhances functional liver tests and reduces hepatotoxicity caused by high doses of paracetamol **(Aller *et al*., 2015)**. Additionally, it has the potential to reduce oxidative stress in rats **(Brandon-Warner *et al*., 2012& Papackova *et al*., 2018).** *Silymarin* displays antiviral and hepatoprotective properties in cases with chronic hepatitis C **(Polyak *et al*., 2013&Lani *et al*., 2015).** Milk thistle exhibits an antibacterial effect and an inhibitory influence on biofilm formation at specific concentrations **(Evren and Yurtcu, 2015).**

Ghaffari *et al*., (2011) discovered that the mean concentrations of AST, ALT, ALP and bilirubin in rats that had methotrexate plus MT were significantly reduced than those that received only methotrexate. The parameters that were compared between the Methotrexate plus milk thistle and control groups (which received only normal saline) weren't statistically significant. Multiple investigations have disclosed the protective impacts of MT in hepatic damages up to this point **(Pradeep *et***

***al*., 2007).** It was demonstrated in these investigations that the treatment period for acute and chronic hepatitis was reduced by the extract of milk thistle **(Saller *et al*., 2007).** Previously, milk thistle was demonstrated to have protective effects in cirrhosis, fatty liver, ischemic liver injury, viral hepatitis and cancer **(Loguercio *et al*., 2007).** Previous research has demonstrated that silymarin inhibits the hepatotoxic impacts of acetaminophen and tetrachloromethane **(Dhiman and Chawla, 2005).** According to reports, MT has the potential to protect the liver from hepatotoxic agents by promoting DNA polymerase, stabilizing all membranes, inhibiting free radicals and increasing glutathione concentration **(Kren and Walterova, 2005).**

Table (6): **The impact of milk thistle extract on serum concentrations of urea nitrogen, ceatinine and uric acid in lead intoxicated rats.**

Parameters / Groups	Urea nitrogen	creatinine	Uric acid
	(mg/dl)		
Group (1) negative control	32.60±0.67^{c,d}	0.76±0.05^{d}	2.76±0.06^{c}
Group (2) positive control	52.60±1.20^{a}	1.82±0.03^{a}	5.96±0.08^{a}
Group (3) 100 mg/kg milk thistle extract	45.40±0.51^{b}	1.50±0.02^{b}	3.32±0.05^{b}
Group (4) 300 mg/kg milk thistle extract	42.40±0.67^{b}	1.45±0.08^{b,c}	2.20±0.03^{d}
Group (5) 500 mg/kg milk thistle extract	38.00±0.83^{c}	0.90±0.04^{c}	3.00±0.03^{b,c}

*Values are expressed as means ±SE.

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

Table (6) illustrates the impact of milk thistle extract on kidney functions, specifically the concentration of urea nitrogen, creatinine and uric acid in the serum. The concentration of serum urea nitrogen was significantly higher in mice exposed to lead toxicity, with an average of 52.60±2.70 (mg/dl), in comparison to the (–ve) group 32.60±1.51 (mg/dl). In contrast to the positive control group, the serum urea nitrogen levels of the rats that were treated with milk thistle extract at any intake level, exhibited a significant decrease to reach normal levels. In comparison to the positive control group 52.60±2.70 (mg/dl), the serum urea nitrogen concentrations of groups three, four, and five were 45.40±1.14(mg/dl), 42.40±1.51(mg/dl) and 38.00±1.87(mg/dl), respectively.

The positive group, which was exposed to the toxicity of lead, exhibited a mean creatinine level of 1.82±0.08 (mg/dl) , which was greater than that of the (–ve) group 0.76±0.11 **(**mg/dl), as

demonstrated in Table (6). The concentration of serum creatinine in the groups of rats that were fed milk thistle extract at any intake level exhibited a decrease in comparison to the +ve group. The optimal level was demonstrated in group five with an average of 0.90±0.10 (mg/dl). In comparison to the -ve group 2.76±0.13 (mg/dl), the (+ve) group exhibited a significant increase in uric acid levels, with a mean value of 5.96±0.19 (mg/dl). Group four (300 mg of MT extract) exhibited a lower value of 2.20±0.08 (mg/dl).

Nouri and Heidarian (2019) stated that rats exhibited a progressive decline in serum uric acid in all treated groups, which was influenced by MT herbs. This result is consistent with their findings. Additionally, **Amin *et al*., (2019)** noted that the uric acid, serum urea and creatinine levels of all acute renal failure groups that were administered varying concentrations of dry milk thistle (20 percent and 40 percent) were significantly lower than those of the control positive group. The biomarkers of renal impairment in the lead-treated groups were significantly different from those in the control group, as indicated by the results of this investigation. This work confirms the findings of **Offor *et al*., (2017)** regarding lead-induced hepato-renal damage in male rats, as we also detected lead-induced renal damage. The findings of this research demonstrate that exposure to lead had a substantial effect on the renal tissues, resulting in a notable increase in the levels of renal damage biomarkers in mice treated with lead, as compared to the control group.

Table (7): **The impact of milk thistle extract on serum concentrations of calcium and phosphorus in lead intoxicated rats.**

Parameters / Groups	Calcium (mg/dl)	Phosphorus (mg/dl)
Group (1) negative control	6.42 ± 0.12^{d}	8.41±0.32^{a}
Group (2) positive control	8.20± 0.28^{b,c}	7.30 ± 0.43^{b}
Group (3) 100 mg/kg milk thistle extract	9.03± 0.38^{a}	6.47±0.41^{c}
Group (4) 300 mg/kg milk thistle extract	8.48± 0.36^{b}	6.08± 0.19 ^{c,d}
Group (5) 500 mg/kg milk thistle extract	8.85± 0.31^{b}	6.45 ± 0.26^{c}

*Values are expressed as means ±SE.

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

The findings in Table (7) demonstrated the impact of milk thistle extract on the concentration of calcium and phosphorus in

the serum. The serum calcium concentration showed a substantial rise in rats subjected to toxicity of lead, with a mean value of 8.20± 0.28 (mg/dl) compared to the (–ve) group with a mean value of 6.42 ± 0.12 (mg/dl). The levels of calcium in serum substantially increased when rat were treated with MT at three different consumption concentrations, with mean values of 9.03±0.38(mg/dl),8.48±0.36(mg/dl)and 8.85±0.31 (mg/dl),respectively. These values were compared to the (+ve) group, which had an average of 8.20± 0.28 (mg/dl).

The data in Table (7) indicated a significant reduction in serum phosphorus concentration when rats were subjected to lead poisoning, with an average of 7.30 ± 0.43 (mg/dl) compared to the negative control group 8.41±0.32 (mg/dl). The serum phosphorus concentrations in rats were significantly reduced when they were fed MT at three different consumption levels. The mean values for the phosphorus concentrations were 6.47±0.41 (mg/dl), 6.08±0.19 (mg/dl) and 6.45±0.26 (mg/dl), respectively. These values were lower compared to the positive control group which had a mean phosphorus concentration of 7.30±0.43 (mg/dl) .

Missoun *et al*., (2010) observed a rise in serum calcium and phosphorus levels in rats that were given lead acetate in their drinking water for a period of eight weeks. This could be attributed to renal dysfunction or the inhibitory impact of lead on tissue transport mechanisms in rats. Furthermore, lead has a direct impact on the functioning of osteoblasts, which includes the inhibition of the synthesis of osteocalcin. Osteocalcin is a significant component of bone that plays a vital role in mineralization. This impact has been shown in an investigation conducted by (**Ronis *et al*., 2001).**

Anetor *et al*., (2005) demonstrated a significant reduction in calcium levels in groups exposed to lead. Hypocalcemia indicates an abnormality in the regulation of calcium levels in the body. The observed elevation in phosphate levels in this research may be attributed to cellular membrane impairment caused by lead exposure. Bucks subjected to eight milligrams of lead acetate/kg b. wt. for four months demonstrated a rise in serum inorganic phosphorus levels, as reported by (**Desouky *et al*., 2001**).

Table (8): The impact of milk thistle extract on serum concentrations of catalase and malondialdehyide in lead intoxicated rats.

Parameters / Groups	CAT (nmol/min/mg protein)	MDA (nmol/min/mg protein)
Group (1) negative control	4.46±0.16[a]	152.20±0.08[e]
Group (2) positive control	3.46±0.08[e]	168.20±0.03[a]
Group (3) 100 mg/kg milk thistle extract	4.04±0.06[d]	91.20±0.06[b]
Group (4) 300 mg/kg milk thistle extract	4.28±0.03[c]	83.40±0.04[c]
Group (5) 500 mg/kg milk thistle extract	4.32±0.05[b]	68.00±0.05[d]

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

The results presented in Table (8) demonstrate the impact of milk thistle extract at various dosages on serum concentrations of antioxidant activity, specifically catalase. The results demonstrated a significant decrease in serum CAT levels in the +ve group, with an average of 3.46±0.18 (nmol/min/mg protein), compared to the (–ve) group 4.46±0.37 (nmol/min/mg protein). Nevertheless, the serum CAT levels exhibited a significant rise ($P < 0.05$) in groups three, four and five when compared to the (+ve) group. Furthermore, the group 5 had the highest value of CAT compared to negative group. However, the results indicated a significant rise in malondialdehyde concentrations in the (+ve) group compare to the (–ve) group. The levels of serum MDA in groups three, four, and five exhibited a significant reduction ($P < 0.05$) compared to the positive group. Furthermore, the five group had the most reduction value of MDA.

Silymarin, a flavonoid derived from the *S. marianum*, can inhibit oxidative stress and may have a beneficial impact on diabetic metabolic abnormalities due to its potent antioxidant properties. Multiple experimental and clinical studies provide evidence supporting the concept that compounds possessing antioxidant characteristics have beneficial effects on the oxidative metabolic dysfunction associated with hyperglycemia **(Packer *et al.*, 2000 & Maddux *et al.*, 2001).**

Several investigations have shown evidence for the antioxidant properties of *silymarin.* For example, it has been observed that *silymarin* can protect against pancreatic damage caused by alloxan by enhancing the activity of antioxidant enzymes for example

GSH and SOD **(Soto, 2003).** In one study, the researchers noted that *silymarin* had a hepatoprotective impact via increasing hepatic GSH levels **(Shaker *et al.*, 2010)**. Furthermore, the administration of *silymarin* demonstrated significant protection of the kidneys by reducing the level of MDA in a model of drug-induced nephrotoxicity **(Guzel, 2019).**

Table (9): **The impact of milk thistle extract on serum concentration of lead in lead intoxicated rats.**

Parameters / Groups	Lead (µg /dl)
Group (1) negative control	211.80±1.06^{e}
Group (2) positive control	462.80±1.15^{a}
Group (3) 100 mg/kg milk thistle Extract	322.80±1.15^{b}
Group (4) 300 mg/kg milk thistle Extract	312.80±1.15^{c}
Group (5) 500 mg/kg milk thistle Extract	282.00±0.83^{d}

*Values are expressed as means ±SE.

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

The results indicated a significant increase in serum lead levels when rats were exposed to lead toxicity (positive control group), with an average of 462.80±2.58 (µg/dL), compared to the negative control group 211.80±2.38 (µg/dL). When rats were given varying amounts of MT in their diet, the pb concentration in their serum declined substantially in all groups. The average values for lead concentration were 322.80±2.50 (µg/dL), 312.80±2.44 (µg/dL) and 282.00±1.87 (µg/dL), respectively compared to the +ve group.

The introduction of oxidative stress caused by lead occurs through the production of reactive oxygen species and the reduction of antioxidant stores **(Elias *et al.*, 2014& Patri *et al.*, 2017)**. The liver plays a crucial role in the metabolism of toxic heavy metals, as it is responsible for their storage, biotransformation, and detoxification **(Aliyu *et al.*, 2015)**. Lead, heavy metal hazardous, induces oxidative damage in the liver by stimulating lipid peroxidation **(Can *et al.*, 2008& Lopes *et al.*, 2016).** According to **Abdel-Moneim (2016)**, lead acetate causes oxidative stress, hepatotoxicity and apoptosis in rats. Previous studies indicate that *silymarin* can have an antioxidant impact through various mechanisms, such as controlling the stability and permeability of cell membranes, regulating intracellular glutathione levels, preventing the transformation of hepatic

stellate cells into myofibroblasts, &stimulating the synthesis of RNA, DNA, & proteins **(Fraschini *et al*., 2002).**

Conclusions

The experimental male albino rats were found to be adversely affected by lead acetate. Thus, this study recommends individuals to consume milk thistle extract (MT) as a preventive measure against potential harmful risks.

References

A.A.C.C. (2000): Approved methods of analysis (10th ed.). *Americ. Associ. of Cere. Chemists. Inc.*

Abdel-Moneim, A. (2016): Indigofera oblongifolia prevents lead acetate induced hepatotoxicity, oxidative stress, fibrosis and apoptosis in rats. *PloS ONE.*, 11(7): e0158965.

Abenavoli, L.; Capasso, R.; Milic, N. and Capasso, F. (2010): Milk thistle in liver diseases: past, present, future. Phytother. Resear.,24(10), 1423- 1432.

Abenavoli, L.; Izzo, A.; Mili´c, N.; Cicala, C.; Santini, A. and Capasso, R. (2018): Milk thistle (Silybum marianum L.): A concise overview on its chemistry, pharmacological, and nutraceutical uses in liver diseases. *Phytother. Res*., 32, 2202–2213.

Abouzid, S.; Ahmed, H.; Moawad, A.; Owis, A.; Chen, S.; Nachtergael, A.; Mcalpine, M.; Friesen, J. and Pauli, G. (2017): Chemotaxonomic and biosynthetic relationships between flavonolignans produced by Silybum marianum populations. *Fitoter*.,119, 175-184

Abouzid, S.; Chen, S. and Pauli, G. (2016): Silymarin content in Silybum marianum populations growing in Egypt. *Indust. crops and produ.*,83, 729-737.

Aebi, H. (1984): Catalase in vitro. In: Methods of Enzymology; 105: 121–126.

Albassam, A.; Frye, R. and Markowitz, J. (2017): The effect of milk thistle (Silybum marianum) and its main

flavonolignans on CYP2C8 enzyme activity in human liver microsomes. *Chemico-biolog. Interact.,* 271, 24-29

Aller, R.; Izaola, O.; Gómez, S.; Tafur, C.; González, G.; Berroa, E. and de Luis, D. (2015): Effect of silymarin plus vitamin E in patients with nonalcoholic fatty liver disease. A randomized clinical pilot study. *Europ. Review for Medic. and Pharmacol. Scien.,*19(16), 3118–3124.

Aliyu, M.; Odunola, O.; Gbadegesin, M.; Adegoke, A.; Olugbami, J. and Ndidi S. (2015): Modulatory role of Acacia honey from North-West Nigeria on sodium arsenite-induced clastogenicity and oxidative stress in male Wistar rats. *Natrl. Prod. Res*., 29(4): 321-26.

Aloia, J.; Vaswani, A.; Yeh, J.; Ellis, K. and Cohn S.(1984): Total body phosphorus in postmenospausal women. Miner Electrolyte *Metab.* ;10:73–76.

Amin, A.; Mosa, Z.; Nagib, E. and Abdelrahman, A. (2019): Effect of feeding artichoke and milk thistle on rats treated with CCl4 and glycerol/saline solution. *Afric. J. of Biologi. Scien*., *15*(1), 1–12.

Anetor, J.; Akingbola, T.; Adeniyi, F. and tylor, G. (2005): Decreased total and ionized calcium levels and haematological indices in occupational lead exposures evidence of the endocrine disruptive effect of lead. *India. J. of Occupati. & Environm. Medic*.,9(1):15-21.

A.O.A.C. (2005): Official Methods of Analysis of the Assoc. of Official Agricultural Chemists, 12th ed. Washington, D.C.

Aslam, S.; Jahan, N.; Ali, S. and Rahman, K. (2012): An innovative microwave- assisted extraction and antioxidant potential of polyphenols from different parts of *Ocimum basilicum*. *J. Med. Plants Res.*, 6, 2150–2159.

Aziz, M.; Saeed, F.; Ahmad, N.; Ahmad, A.; Afzaal, M.; Hussain, S.; Mohamed, A.; Alamri, M. and Anjum, F. (2021): Biochemical profile of milk thistle *(Silybum Marianum L.)* with special reference to silymarin content. *Food Sci. Nutr*., 9, 244–250.

Biedermann, D.; Vavˇríková, E.; Cvak, L. and Kˇren, V. (2014): Chemistry of silybin. *Nat. Prod. Rep.*,31, 1138–1157.

Bijak, M. (2017): Silybin, a Major Bioactive Component of Milk Thistle (Silybum marianum L. Gaernt.)—Chemistry, Bioavailability, and Metabolism. *Molecul*., 22, 1942.

Brandon- Warner, E.; Eheim, A.; Foureau, D.; Walling, T.; Schrum, L. and McKillop, I. (2012): Silibinin (Milk Thistle) potentiates ethanoldependent hepatocellular carcinoma progression in male mice. *Cancer Lette*.,326(1), 88–95.

Burstein, M. (1970): Rapid method for isolation of lipoproteins from human serum by precipitation with poly-anion. *J. lipid Resear*., 11: 583- 588.

Can, S.; Bagci, C.; Ozaslan, M.; Bozkurt, A.; Cengiz, B.; Cakmak, E.; Kocabaş, R.; Karadag, E. and Tarakçioğlu. M. (2008): Occupational lead exposure effect on liver functions and biochemical parameters. *Acta. Physiol. Hung*.,95: 395-403.

Chambers, C.; Holeˇcková, V.; Petrásková, L.; Biedermann, D.; Valentová, K.; Buchta, M. and Kˇren, V. (2017): The *silymarin* composition and why does it matter? *Food Res. Int*., 100, 339–353.

Cintra, D.; Costa, A.; Peluzio, M.; Matta, S.; Silva, M. and Costa, N. (2006): Lipid profile ofrats fed high fat diets based on flaxseed peanut, trout or chicken skin. *Nutr.*, 22: 197-205.

Dabbour, I.; Ahmad, M.; Takruri, H. and Al- Ismail, K. (2014): Effect of chemical properties of milk thistle seed oil on serum lipid profile and antioxidants capacity in rats fed high cholesterol and cholesterol free diets. *Pakis. J. Nutr*. 13(10): 600-609.

Desouky, H.; El-Sheshtawy, R.; Hanafi, E.; Shalaby, S. and Ahmed, W. (2001): GnRH response in lead exposed baladi bucks with emphasis on seminal, blood and

testicular alterations. *J. of Egyptian Veterin. Medic. Associa*.,61:127-142.

Detaille, D.; Sanchez, C.; Sanz, N.; Lopez-Novoa, J.; Leverve, X. and El-Mir, M. (2008): Interrelation between the inhibition of glycolytic flux by silibinin and the lowering of mitochondrial ROS production in perifused rat hepatocytes. *Life Sci.*,82(21-22):1070–6.

Dhiman, R. and Chawla, Y. (2005): Herbal medicines for liver diseases. *Dig Dis Sci.*,50(10):1807–12.

Draper, H. and Hadley, M. (1990): Malondialdehyde determination as index of lipid peroxidation. *Metho. Enzymol.*, 186, 421-431.

Drouet, S.; Abbasi, B.; Falguières, A.; Ahmad, W.; Sumaira Ferroud, C.; Doussot, J.; Vanier, J.; Lainé, É. and Hano, C. (2018): Single Laboratory Validation of a Quantitative Core Shell-Based LC Separation for the Evaluation of *Silymarin* Variability and Associated Antioxidant Activity of Pakistani Ecotypes of Milk Thistle *(Silybum marianum L.). Molecul.*,23, 904.

El-Elimat, T.; Raja, H.; Graf, T.; Faeth, S.; Cech, N. and Oberlies, N. (2014): Flavonolignans from Aspergillus iizukae, a Fungal Endophyte of Milk Thistle (Silybum marianum L.). *J. Nat. Prod*., 77, 193–199.

Elias, A.; Oputiri, P.; Oru-Bo, A. and Enimeya. A. (2014): Lead Organ and Tissue Toxicity: Roles of Mitigating Agents (Part 2). *British. J. Pharmacol. Toxicol*., 5(1): 1-15.

Evren, E. and Yurtcu, E. (2015): In vitro effects on biofilm viability and antibacterial and antiadherent activities of silymarin. *Foli. Microbiol.*, 60(4), 351–356.

Federico, A.; Dallio, M. and Loguercio, C. (2017): Silymarin/silybin and chronic liver disease: A marriage of many years. *Molecul.*,22(2), pii: E191.

Flora, S. (2009): Structural, chemical and biological aspects of antioxidants for strategies against metal and metalloid exposure. *Oxid. Med. Cell Longev.*, 2: 191–206.

Fossati, P.; Prencipe, L. and Berti, G. (1980): Enzymatic colorimetric method of determination of urea in serum. Clin.*Chem*., 6(18) 499-502.

Fossati, P. and Prencie, L. (1982): Serum triglycerides determined colorimeterically with an enzyme that produces hydrogen peroxide. *Clin. Chem*., 28:2077-2080.

Fraschini, F.; Demartini, G. and Esposti, D. (2002): Pharmacology of silymarin. *Clin. Drug Investig*., 22: 51–65.

Friedewald, T.; Levy, R. and Fredrichsor, D. (1972): Estimation of the concentration of low-density lipioprotein cholesterol in plasma without use of the preparative ultracentrifuge. *Clin. Chem*., 18:499-502.

Gagan, F.; Deepesh, G. and Archana. T. (2012): Toxicity of lead: A review with recent updates. *Interdiscip. Toxicol.*, 5(2): 47-58.

Ghaffari, A.; Noshad, H.; Ostadi, A.; Ghojazadeh, M. and Asadi, P. (2011): The effects of milk thistle on hepatic fibrosis due to methotrexate in rat. *Hepat. Mon*.11(6):464-8.

Gorinstein, S.; Leontowicz, A.; Lojek, M.; Ciz, Krzminski, R.; Zachwieja, Z.; Jastrzebski, Z.; Delgado-Licon, E.; Martin Belloso, O. and Trachtenberg, S. (2003): Seed oil improve lipid metabolism and increase antioxidant potential in rats fed diets containing cholesterol. *Nutr. Res*., 23: 317-330

Guzel, S. (2019): Potential renoprotective effects of silymarin against vancomycin-induced nephrotoxicity in rats. *Drug Chem. Toxicol.*, p. 1-7.

Henry, R. (1974): Creatinine measurement with colorimetric method. In clinical Chem., Principles and technics.

Second edition, Haper and Row puplishers. hepatocytes. *Cancer Lett*., 97: 61-67.

Henry, R.; Cannon, D. and Winkelman, J. (1974): Clinical Chemistry Principles and Techniques, Harper and Row. New York, pp:1440-1452.

Hwang, D. and Wang, L. (2001): Effect of taurine on toxicity of cadmium in rats. *Toxicolo.*,167:173–180.

Ibrahim, N.; Eweis, E.; El-Beltagi, H. and Abdel-Mobdy, Y. (2012): Effect of lead acetate toxicity on experimental male albino rat. *Asian Pac. J. Trop. Biomed.*, 2(1):41-6.

Javeed, A.; Ahmed, M.; Sajid, A.; Sikandar, A.; Aslam, M.; Hassan, T.; Samiullah Nazir, Z.; Ji, M. and Li, C. (2022): Comparative Assessment of Phytoconstituents, Antioxidant Activity and Chemical Analysis of Different Parts of Milk Thistle *Silybum marianum L. Molecu*.**,** 27, 2641.

Kim, J.; Na Lee, Y.; Lee, J.; Lee, S.; Kim, H.; Choi, Y.; Draelos, Z. and Kim, J. (2023): Efficacy and safety of *silymarin* containing antioxidant serum as an adjuvant treatment of mild-to-moderate acne vulgaris: A prospective, open-label pilot study. *J. Cosmet. Dermatol*.**,** 22, 561–568.

Kren, V. and Walterova, D. (2005): Silybin and silymarin--new effects and applications. *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub.*,149(1):29–41.

Kumar, S. and Khanna, R. (2018): Beneficial effects of *Silybum marianum* seed extract against hepatic fibrosis induced by carbon tetrachloride in rats. *B.M.J. J.* 67, (2): AB.CO 265.

Landrigan, P. and Fuller. R. (2015): Environmental pollution: an enormous and invisible burden on health systems in low- and middle-income countries. *World Hosp. Heal. Serv.*, 50(4): 35-41.

Lani, R.; Hassandarvish, P.; Chiam, C.; Moghaddam, E.; Chu, J.; Rausalu, K. and Zandi, K. (2015): Antiviral

activity of silymarin against chikungunya virus. Scienti. Repo.,5, 11421.

Loguercio, C. (2011): Silybin and the liver: From basic research to clinical practice. *World J. Gastroenterol*.,17, 2288–2301.

Loguercio, C.; Federico, A.; Trappoliere, M.; Tuccillo, C.; de Sio, I.; Di Leva, A.; Niosi, M.; D'Auria, M.; Capasso, R. and Del Vecchio Blanco, C (2007): The effect of a silybin-vitamin e-phospholipid complex on nonalcoholic fatty liver disease: a pilot study. *Dig Dis Sci.* 2007;52(9):2387–95.

Lopes, A.; Peixe, T.; Mesas, A. and Paoliello. M. (2016): Lead exposure and oxidative stress: A systematic review. In : de Voogt P. (eds). *Rev. Environ. Contam. Toxicol.*, Volume 236 : 193-238.

MacDonald-Ramos, K.; Michán, L.; Martínez-Ibarra, A. and Cerbón, M. (2020): *Silymarin* is an ally against insulin resistance: *A review*. *Ann. Hepatol*., 23, 100255.

Maddux, B.; See, W.; Lawrence, J.; Goldfine, A.; Goldfine, I. and Evans J. (2001): Protection against oxidative stress-induced insulin resistance in rat L6 muscle cells by mircomolar concentrations of alpha-lipoic acid. *Diabetes*.50 (2):404–410.

Makni, M.; Fetoui, N.; Gargouri, M.; Garoui, E.; Jaber, H.; Makni, J.; Boudawara T. and Zeghal, N. (2009): Hypolipdemic and hepatoprotective effects of flax and pumpkin seed mixture rich in n-3 and n-6 fattya cids in hypercholesterolemic rats. *Food Chem. Toxicol.*, 46: 3714-3720.

Marceddu, R.; Dinolfo, L.; Carrubba, A.; Sarno, M. and Di Miceli, G. (2022): Milk Thistle *(Silybum marianum L.*) as a Novel Multipurpose Crop for Agriculture in Marginal Environments: *A Review*. *Agron.*,12, 729.

Marin, V.; Gazzin, S.; Gambaro, S.; Dal, B.; Calligaris, S.; Anese, M. and Rosso, N. (2017): Effects of oral

administration of silymarin in a juvenile murine model of non- alcoholic steatohepatitis. *Nutrien.*, 9(9), 1006.

Medecins San Frontieres (MSF). (2012): Lead Poisoning Crisis in Zamfara State, Northern Nigeria. MSF Briefing Paper. 4.

Missoun, F.; Slimani, M. and Aoues, A. (2010) Toxic effect of lead on kidneyfunction in rat wistar. *African J. of Biochem.Research*,4(2):21-27.

Moon, K.; Back, S.; Kim, J.; Jean, S. and Choi, M. (2001) Safflower seed extract lowers plasma and hepatic lipids in rats fed high-cholesterol diet. *Nutr. Res*., 21: 895-904.

Nouri, A. and Heidarian, E. (2019): Nephroprotective effect of *silymarin* against diclofenac-induced renal damage and oxidative stress in male rats. *J. of Herb: Med. Pharmacol*., *8*(2), 146–152.

Newairy, A. and Abdou, H. (2009): Protective role of flax lignans against lead acetate induced oxidative damage and hyperlipidemia in rats. *Food Chem. Toxicol*., 47(4):813-818.

Ni, X. and Wang, H. (2016): *Silymarin* attenuated hepatic steatosis through regulation of lipid metabolism and oxidative stress in a mouse model of nonalcoholic fatty liver disease (NAFLD). *Americ. J. of translate. l resear*., 8(2): 1073.

Offor, S.; Mbagwu, H. and Orisakwe. O. (2017): Lead induced hepato-renal damage in male albino rats and effects of activated charcoal. *Front Pharmacol*., 8:107.

Packer, L.; Rosen, P.; Tritschler, H.; King, G. and Azzi, A. (2000): Antioxidants and diabetes management. New York: Marcel Dekker.

Papackova, Z.; Heczkova, M.; Dankova, H.; Sticova, E.; Lodererova, A.; Bartonova, L. and Cahova, M. (2018): Silymarin prevents acetaminophen- induced hepatotoxicity in mice. *PLoS ONE*., 13(1), e0191353.

Patra, R.; Rautray A. and Swarup, D. (2011): Oxidative stress in lead and cadmium toxicity and its amelioration. *Vet. Med. Int.*, 1-9.

Patri, S.; Sahu S.; Parida, B.; Baral, Prusty, A.; Samanta, L. and Jena. S. (2017): Effect of Lead acetate on oxidative stress and antioxidant defense system of Bacillus subtilis and plasmid (pBSIISK) isolated from DH5α . In: Abstracts of the NGBT conference; Bhubaneswar, Odisha, *Indi.Can. J. Biotech*. 1: 154.

Parsons, P. (2001): Analytical procedures for the determination of lead in blood and urine; approved guideline. *Wayne, PA, National Committee for Clinical Laboratory Standards*. 113(8):1669–1674.

Pellet, P. and Sossy, S. (1970): Food composition. Tables for use in the Middle East. *American Univ. of Beirut. Beirut. Lebanon*., 2:126.

Polyak, S.; Ferenci, P. and Pawlotsky, J. (2013): Hepatoprotective and antiviral functions of silymarin components in HCV infection. *Hepatology*, 57(3), 1262–1271.

Porupa, M.; Matuskova, Z.; Kazdova, L.; Oliyarnyk, O.; Malinska, H.; di Angelo, I.T. and Vecera, R. (2015). Positive effects of different drug forms of silybin in the treatment of metabolic syndrome. *Physiolo. Res*., 64: 507.

Pradeep, K.; Mohan, C.; Gobianand, K. and Karthikeyan, S. (2007): Silymarin modulates the oxidant-antioxidant imbalance during diethylnitrosamine induced oxidative stress in rats. *Eur. J. Pharmacol.*, 560(2-3):110–6.

Raclariu-Manolic˘a, A. and Socaciu, C. (2023): Detecting and Profiling of Milk Thistle Metabolites in Food Supplements: A Safety-Oriented Approach by Advanced Analytics. *Metabol*., 13, 440.

Reeves, R.; Nielsen F. and Fahey G. (1993): AIN-93 Purified Diets for Laboratory Rodents .*J. Nutr*.,123(1):1939-1951.

Ronis, M.; Aronson, J.; Gao, G.; Hogue, W .; Skinner, R.; Badger,T. and Lumpkin, C. (2001): Skeletal Effect of developmental lead exposure in rats. *Toxicol. Sci*., 62:321-329.

Roy, E., (1970): Colorimetric determination of Co. St Louis. Toronto. Princeton; 1088-1273.

Ross, S.(2008): Milk thistle (Silybum marianum): an ancient botanical medicine for modern times. *Holist. Nurs. Pract.*, 22(5):299–300.

Saller, R.; Melzer, R. and Brlgnoli, R. (2001): The use of silymarin in the treatment of liver diseases. Drugs, 61(14), 2035-2063.

Saller, R.; Melzer, J.; Reichling, J.; Brignoli, R. and Meier, R. (2007): An updated systematic review of the pharmacology of silymarin. *Forsch. Komplementmed.*,14(2):70–80.

Seddik, L.; Bah, T.; Aoues, A.; Brnderdour, M. and Silmani, M. (2010): Dried leaf extract protects against lead-induced neurotoxicity in Wistar rats. *Eur. J. Sci. Res.*, 42(1):139–151.

Shaker, E.; Mahmoud, H. and Mnaa, S. (2010): Silymarin, the antioxidant component and Silybum marianum extracts prevent liver damage. *Food and Chemical Toxico*., 48(3): p. 803-806

Sherwin, J. (1984): Liver Function. In Kaplan LA, Pesce AJ, eds. Clinical chemistry, theory, analysis, and correlation. St Louis: Mosby 55(25):420-438.

Schultz, A.(1984): Uric Kaplan A. Clin Chem. Mosby Co. St. Louis Toronto. Princeton; 1261-1266 and 41**.**

Siegel, A. and Stebbing, J. (2013): Milk thistle: Early seeds of potential. Lancet Oncol. 14, 929–930.

Snedecor, G. and Cochran, W. (1980): Statistical methods.,7th Ed., Iowa State University Press, *Ames, USA.*, (90).

Soliman, S.; Sherif, H.; Bonfill, M.; EL-Garhy, H. (2018):

Bioinformatics and differential expression analysis of chalcone synthase genes (CHS1, 2, 3) under gamma rays elicitation in Silybum marianum L. Conference: 4th International Conference on Biotechnology Applications in Agriculture (ICBAA), *Benha UniversityAt: Hurghada, Egypt.* 1-11.

Soto, C.(2003): Silymarin increases antioxidant enzymes in alloxan-induced diabetes in rat pancreas. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology., 136(3): p. 205-212.

Sunday, A.; Iliyasu, O.; Wilson, O.; Augustine, E. and Uduak, U. (2012): Preventive activity of ascorbic acid on lead acetate induced cerebellar damaged in adult wistar rats. *Open Access Res. J.* 13: 99-104.

Tsai, J.; Liu, J.; Wu, T.; Ho, P.; Huang, C.; Shyu, J.; Hsieh, Y.; Tsai, C. and Liu, Y.(2008): Effects of silymarin on the resolution of liver fibrosis induced by carbon tetrachloride in rats. *J. Viral. Hepat.*,15(7):508–14.

Vargas- Mendoza, N.; Madrigal- Santillán, E.; Morales-González, A.; Esquivel- Soto, J.; Esquivel- Chirino, C.; García- Luna, y.; González- Rubio, M. and Morales- González, J. (2014): Hepatoprotective effect of silymarin. *World J. of Hepatol*., 6(3), 144–149.

Victor, C.; Rippon, I.; Nelis, S.; Martyr, A.; Litherland, R.; Pickett, J.; Hart, N.; Henley, J.; Matthews, F.and Clare L.(2020): Prevalence and determinants of loneliness in people living with dementia: Findings from the IDEAL programme. *Int .J. Geriatr Psychiatry*. ;35(8):851-858.

Visavadiya, N. and Narasimhacherya, A. (2008): Sesame as a hypocholestraemic and antioxidant. dietary component. *Food Chem. Toxicol.*, 46: 1889- 1895.

Young, D. (1990): Effect of drugs on clinical laboratory tests. *Am. J. Clin. Pathol*., 3(7):6-12.

تأثير مستخلص حليب الشوك على التسمم بالرصاص فى الفئران

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الملخص العربى

كان الهدف من اجراء هذا البحث هو معرفة التأثير الغذائي لمستخلص حليب الشوك وفاعليته ضد التسمم بالرصاص في الفئران. استخدمت التجربة 30 فأر من ذكور الالبينو تزن 140±5جم، مقسمين إلى خمس مجموعات (6 فئران لكل مجموعة). المجموعة الأولى مجموعه ضابطة سالبة، والثانية مجموعه ضابطة موجبة تغذت على غذاء مضاف إليه 200 ملجم/كجم خلات الرصاص. المجموعات (3–5) تناولت الغذاء الأساسي مضافًا إليه نفس كمية خلات الرصاص بالإضافة إلى 100 ملجم، 300 ملجم، و500 ملجم/كجم من مستخلص حليب الشوك على التوالي لمدة ستة أسابيع. تم تقدير التركيب الكيميائي لحليب الشوك، معدل الزيادة في الوزن، ومستويات الكالسيوم، الفوسفور، الرصاص، وظائف الكلى والكبد، المالوندهيد وبعض الأنزيمات المؤكسدة.

أظهرت النتائج زيادة في وزن الفئران التي تغذت على مستخلص حليب الشوك مقارنة بالمجموعة الضابطة الموجبة. كما انخفضت أنزيمات الكبد والكلى في المجموعات (3–5) مقارنة بالمجموعة الضابطة الموجبة. تحسنت مستويات الكالسيوم والفوسفور في المجموعات التي تغذت على المستخلص. وانخفضت مستويات الرصاص في هذه المجموعات. وزادت مستويات الكتاليز في المجموعات (3–5) بينما انخفض المالوندهيد. يوصي البحث بأن استهلاك مستخلص حليب الشوك له تأثير إيجابي على الصحة عبر تقليل التأثير الضار الناتج من الرصاص.

الكلمات المفتاحية: بلسم الليمون، الانزيمات المؤكسدة، الالتهاب الكلوى الحاد، وظائف الكبد.