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جامعة عين شمس، كلية التربية النوعية، القاهرة، مصر

تحية طيبة وبعد،،،

يسر **معامل التأثير والاستشهادات المرجعية للمجلات العلمية العربية (ارسيف – ARCIF)**، أحد مبادرات قاعدة بيانات **"معرفة"** للإنتاج والمحتوى العلمي، إعلامكم بأنه قد **أطلق التقرير السنوي الثامن للمجلات للعام 2023.**

ويسرنا تهنئتكم وإعلامكم بأن **المجلة المصرية للدراسات المتخصصة** الصادرة عن **جامعة عين شمس، كلية التربية النوعية، القاهرة، مصر، قد نجحت في تحقيق معايير اعتماد معامل "ارسيف Arcif"** المتوافقة مع المعايير العالمية، والتي يبلغ عددها **(32) معياراً**، وللاطلاع على هذه المعايير يمكنكم الدخول إلى الرابط التالي:

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* **بحوث علمية محكمة باللغة الإنجليزية :**

The efficacy of some foods in reducing some heavy metal accumulation in rat's liver

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The efficacy of some foods in reducing some heavy metal accumulation in rat's liver

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Abstract

Lead (Pb), cadmium (Cd), and mercury (Hg) are heavy metals that can cause strong biological effects. Some foods can neutralize or detoxify toxins and protect the liver from the toxic effects of heavy metals. These foods included garlic, ginger, carrot, corn, aloe vera, honey, milk thistle and Dates. The main objective of the present study was to indorse specific food formulas as detoxifying to minimize the harmful effect of lead, cadmium, and mercury. In this study, we investigated the effect of some foods on the toxicity of cadmium chloride (150 mg/l), mercury chloride (80 mg/l) and lead acetate (160 mg/l) in drinking water for 4 weeks on liver function of male rats. The results indicated that garlic, ginger, and milk thistles (formula 1) and garlic, ginger, carrot, corn, date mesocarp (pulp), aloe vera, and honey (formula 2) decrease the activity of the serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) and malondialdehyde (MDA) While increase Catalase (CAT) activity. Also (formula 2) recorded the best results compared with (formula 1). The pathological study of the tissues showed a complete improvement in the liver tissues after lead, cadmium and mercury caused severe liver damage. Concomitant use of (formula 2) and (formula 1) was found to reduce lead, cadmium and mercury concentration considerably indicating the potential therapeutic activity of (formula 2) and (formula 1) against heavy metals toxicity.

Keywords: Detaxation, Medical plants, Garlic, Ginger, Carrot, Corn, Date, Milk thistles, Lead, Cadmium, Mercury, and Heavy metals

Introduction

The pollution of the environment caused by dyes and heavy metals emitted by industries has become a worldwide problem. (Singh et al., 2022) Heavy metals can be transferred and biomagnified via food chains and seriously threaten human health (Zaynab et al., 2022). Lead, cadmium and mercury are toxic metals that are widespread in the environment from natural and

anthropogenic sources. Lead is transferred freely across the placenta (the ratio of fetal:maternal blood lead is about 0.7–0.9 and across the blood-brain barrier, as is mercury; transfer of cadmium is less marked but it tends to accumulate in the placental tissue where it may interfere with zinc transport and affects endocrine hormone synthesis and cellular functions. (Taylor, et al., 2018) Toxic manifestations of these metals are attributed primarily to oxidative stress (Flora et al., 2008). Oxidative stress is defined as an imbalance between the production of free radicals and reactive metabolites, so-called oxidants, and their elimination by antioxidant systems. This imbalance leads to damage to important biomolecules and organs with a potential impact on the whole organism (Duracková, 2010). The associated DNA, protein, and lipid damage may underlie liver diseases as a key pathophysiological force. The above may also be related to chronic liver injury, hepatic inflammation, fibrosis, and hepatocellular carcinoma (Vera-Ramirez et al., 2013) The liver is an important organ to be considered when the effects of pollutants are investigated, since this organ plays a central role in the metabolism and detoxification of biological substances. Also, most of the substances absorbed by the intestine pass first through the liver where toxins and heavy metals may accumulate (Saïdi et al., 2013). Therefore, it was necessary to detoxification of those heavy metals, Detoxification is the process of purifying

the body from compounds that have a detrimental effect on cell functions or structures. Modern research has shown that a wide range of plants can neutralize or detoxify toxins and protect respiratory, urinary, hepatic, and neural systems from the toxic effects of drugs and chemicals (Al-Snafi, 2015). One of these plants is garlic, it shows a protective effect against heavy metals poisoning in mice and co-administration of garlic with cadmium or organic mercury for 12 weeks reduces the accumulation of heavy metals in the liver, kidneys, bones, and testicles. (Senapati et al., 2019). In addition, the simultaneous consumption of milk thistle, results in more improved detoxification of lead in rats'

livers. In patients with liver problems, milk thistle could improve liver function (Wellington and Jarvis, 2019). Also, ginger has detoxifying and antioxidant effects by many phenolic antioxidant compounds that may play part in protecting the body against the hazard effects (Abdel-Gabbar et al., 2019). Also, histopathological studies confirmed the curative efficacy of the extract of aloe vera liver damage (Kumar et al., 2019). The main objective of the present study was to indorse specific food formulas as detoxifying to minimize the harmful effect of lead, cadmium, and mercury.

Martials and Methods

Martials

Garlic, Carrot, Ginger, and Corn were purchased from the local market, in Cairo, Egypt, but aloe vera (L.) Brum family (Liliaceae) was obtained from the Orman-botanic-garden in Giza, Egypt, also Dates palms were obtained from altahhan dates, Cairo, Egypt, and Honey was obtained from Alaseal Honey, in Cairo, Egypt. Samples collections were conducted during the months of May and June 2021.

Lead, Cadmium and Mercury were obtained from Nile Pharmaceutical Company Cairo, Egypt.

Preparation of dried plants

Fresh Garlic, Ginger, Carrot, Corn, Aloe Vera, and Dates were cleaned and washed also dried in an oven under vacuum Then it is ground They were stored in tightly sealed dark containers in a freezer at −1°C for later use.

Biological experiment

Animal, housing, and diets:

Sixty-five male albino rats weighing about 220 ±15 g were obtained from the Agricultural Research Center, Giza, Egypt. The animals were randomly divided into thirteen groups of five rats each.

Group one of rats control negative was fed on basal diet for 8 weeks (total period of experimental). Group two (control positive) received daily feeding of drinking water containing lead chloride (160 mg/l) for four weeks (Sajitha et al., 2010) and fed on a basal diet. Groups three and four received daily feeding of drinking water containing lead chloride (160 mg/l) for four weeks like group 2 then fed on formula (1) containing (1.8g garlic + 2g Ginger+ 0.5g milk thistle) and formula (2) contain (0.9g garlic + 1g Ginger+ 3g corn+ 3.5g dates+ 2.5g Carrots+ 0.1g Aloe Vera), respectively mixed with the basal diet for remnant four weeks. Group five (control positive (was consumed a solution of cadmium chloride (CdCl2) (150 mg/l) as drinking water for four weeks (Haouem et al., 2013) with fed on a basal diet. Groups six and seven consumed a solution of cadmium chloride (CdCl2) (150 mg/l) as drinking water for four weeks like group five then fed on formula (1) and formula (2), respectively mixed with the basal diet for remnant four weeks. Group eight (control positive) consumed a solution of mercury chloride (HgCl2) (80 mg/l) as drinking water for four weeks (Haouem et al., 2013) with fed on a basal diet. Groups nine and ten were consumed a solution of mercury chloride (HgCl2) (80 mg/l) as drinking water for four weeks like group eight then fed on formula (1) and formula (2) Respectively mixed with the basal diet for remnant four weeks.

Group eleven (control positive) consumed a solution from a mixture of lead chloride (160 mg/l), cadmium chloride (150 mg/l) and mercury chloride (80 mg/l) as drinking water for four weeks (Sajitha et al., 2010) and (Haouem et al., 2013) with fed on basal diet were consumed a solution of mercury chloride (HgCl2) (80 mg/l) as drinking water for four weeks like group eight then fed on formula (1) and formula (2) Respectively mixed with the basal diet for remnant four weeks. Group Eleven (control positive) consumed a solution from a mixture of lead chloride (160 mg/l), cadmium chloride (150 mg/l) and mercury chloride (80 mg/l) as drinking water for four weeks (Sajitha et al., 2010) and (Haouem et al., 2013) with fed on a basal diet. Group Twelve and Thirteen

consumed a solution from a mixture of lead chloride (160 mg/l), cadmium chloride (150 mg/l) and mercury chloride (80 mg/l) as drinking water for four weeks like group eleven then fed on formula (1) and formula (2), respectively mixed with the basal diet for remnant four weeks.

The following steps by Schemer (1967) were done in rats after six weeks of treatment in each group

- The animals fasted for 12 h.
- Blood samples were withdrawn from orbital plexus venous by using fine capillary glass tubes.
- Blood samples were collected into plain tubes without anticoagulant
- Blood samples were centrifuged at 3000 rpm for 10 min at 4°C, to obtain clear serum.
- Serum was frozen at -18oC until analyzed.
- The animals were anesthetized with ether and sacrificed.
- They were quickly dissected to excise the liver.
- Liver was weighed and then kept until histological investigations.

Histopathology Technique

The tissue sample from the liver and kidney were fixed immediately after dissection in 10% neutral formalin for 24 hours, then collected and dehydrated using ascending grades of alcohol, cleaned in xylene, and embedded in paraffin wax. Tissues were sectioned at a thickness of 5 microns and stained with hematoxylin and eosin (Banchroft et al., 1996). Then examined by the light microscope for detection of any histopathological alteration.

Biochemical analysis

Blood samples were withdrawn from orbital plexus venous by using fine capillary glass tubes, placed in centrifuge tubes without anticoagulant and allowed to clot. After the serum was prepared by centrifugation (3000 rpm for 15 min), serum samples were analyzed by bio diagnostic kits.

- Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities were determined according to Henry et al (1960).
- Catalase (CAT) activity was determined according to Johansson and Borg (1988).
- Malondialdehyde (MDA) was determined according to Ohkawa et al., (1979).

Statistical Analysis

The obtained data were exposed to the analysis of variance. Duncan's multiple range test at a 5% level of significance was used to compare between means. The analysis was carried out using the PROC ANOVA procedure of the Statistical Analysis System (SAS, 2006).

RESULTS AND DISCUSSION

Biological evaluation of some foods on experimental rats

No rats among the groups died during the experimental period (8 weeks) and all the rats in the groups exhibited no abnormal signs throughout the test period.

Biochemical analysis

Results of liver enzymes ALT and AST of all tested groups are presented in Table (1). Alterations in ALT and AST after 4 weeks post-treatment with lead in the positive control group (78.56±1.02 and 81.20±0.95) there were statistically significant ($P \leq 0.05$) elevations in ALT and AST when compared with negative control groups (36.03±1.51 and 43.08±0.93) (which was fed the

experimental diets). Results of ALT and AST activity after 4 weeks post-treatment with lead then 4 weeks of treatment with different combination of plants in the treatment groups (59.85±1.28 and 69.25±2.00) formula (1) and (52.58±2.43 and 62.39±0.49) formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (ALT and ALT) when compared with positive control groups (78.56±1.02 and 81.20±0.95). There was a significant ($P \leq 0.05$) decrease in the activities of (ALT and AST) between formula (1) and formula (2) in lead fed groups in favor of formula (2)

Table (1): Liver function of experimental rats treated with different combinations of some food (formula 1 and formula 2) after induced toxicity with heavy metals

parameters	Control (-)	Control (+)	Treated groups (U/ml)	
			Formula (1)	Formula (2)
Lead -fed groups				
ALT	36.03±1.51^{d}	78.56±1.02^{a}	59.85±1.28^{b}	52.58±2.43^{c}
AST	43.08±0.93^{d}	81.20±0.95^{a}	69.25±2.00^{b}	62.39±0.49^{c}
Cadmium -fed groups				
ALT	36.03±1.51^{d}	86.51±3.97^{a}	75.33±1.18^{b}	61.56±1.98^{c}
AST	43.08±0.93^{d}	94.03±0.49^{a}	86.91±0.66^{b}	64.28±0.76^{c}
Mercury -fed groups				
ALT	36.03±1.51^{d}	86.31±1.84^{a}	79.30±1.29^{b}	67.86±1.34^{c}
AST	43.08±0.93^{d}	97.24±0.99^{a}	85.57±0.56^{b}	72.27±1.26^{c}
Lead, Cadmium and Mercury -fed groups				
ALT	36.03±1.51^{d}	103.35±1.79^{a}	87.02±2.44^{b}	74.06±2.48^{c}
AST	43.08±0.93^{d}	118.11±2.90^{a}	93.56±2.09^{b}	84.85±3.31^{c}

* Data are presented as means ± SDM.

* Data in a row with different superscript letters are statistically different ($P \leq 0.05$).

AST: aspartate amino transferase ALT: alanine amino transferase

Results of liver enzymes ALT and AST of all tested groups are presented in Table (1). Alterations in ALT and AST after 4 weeks post-treatment with cadmium in the positive control group (86.51±3.97 and 94.03±0.49) there were statistically significant ($P \leq 0.05$) elevations in ALT and AST when compared with negative control groups (36.03±1.51 and 43.08±0.93) (which was fed the

experimental diets). Results of ALT and AST activity after 4 weeks post-treatment with cadmium and then 4 weeks of treatment with different combination of plants in the treatment groups (75.33±1.18 and 86.91±0.66) formula (1) and (61.56±1.98 and 64.28±0.76) formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (ALT and ALT) when compared with positive control groups (86.51±3.97 and 94.03±0.49). There was a significant ($P \leq 0.05$) decrease in the activities of (ALT and AST) between formula (1) and formula (2) in cadmium-fed groups in favor of formula (2). Results of liver enzymes ALT and AST of all tested groups are presented in Table (1). Alterations in ALT and AST after 4 weeks post-treatment with mercury in the positive control group (86.31±1.84 and 97.24±0.99) there were statistically significant ($P \leq 0.05$) elevations in ALT and AST when compared with negative control groups (36.03±1.51 and 43.08±0.93) (which was fed the experimental diets). Results of ALT and AST activity after 4 weeks post-treatment with mercury and then 4 weeks of treatment with different combinations of plants in the treatment groups (79.30±1.29 and 85.57±0.56) formula (1) and (67.86±1.34 and 72.27±1.26) formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (ALT and ALT) when compared with positive control groups (86.31±1.84 and 97.24±0.99). There was a significant ($P \leq 0.05$) decrease in the activities of (ALT and AST) between formula (1) and formula (2) in mercury-fed groups in favor of formula (2).

Results of liver enzymes ALT and AST of all tested groups are presented in Table (1). Alterations in ALT and AST after 4 weeks post-treatment with a mixture of lead, cadmium, and mercury in the positive control group (103.35±1.79 and 118.11±2.90) there were statistically significant ($P \leq 0.05$) elevations in ALT and AST when compared with negative control groups (36.03±1.51 and 43.08±0.93) (which was fed the experimental diets). Results of ALT and AST activity after 4 weeks post-treatment with a mixture of lead, cadmium and mercury then 4 weeks of treatment with different combination of

plants in the treatment groups (87.02±2.44 and 93.56±2.09) formula (1) and (74.06±2.48 and 84.85±3.31) formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (ALT and ALT) when compared with positive control groups (103.35±1.79 and 118.11±2.90). There was a significant ($P \leq 0.05$) decrease in the activities of (ALT and AST) between formula (1) and formula (2) in a mixture of lead, cadmium, and mercury-fed groups in favor of formula (2).

There is an association between hepatotoxicity and blood lead levels. (Nakhaee et al., 2019). It has been shown that Lead exposure is linked to perturbations of synthetic liver function and elevations in liver biomarkers such as alanine aminotransferase and aspartate aminotransferase. (Reja et al., 2020) The accumulation of significant amounts of lead in liver tissue was implicated in the induction of an oxidative stress response in the liver. (Suleiman et al., 2013) Attention has been developed on the protective biochemical function of the natural antioxidants contained in the dietary plants that are candidates for prevention or protection of oxidative damage caused by free radicals' species. (Victoria et al., 2004). This improvement in liver enzymes is due to the curative properties of garlic, ginger, milk thistle, carrots, corn, dates, and honey, which our results agreed with the results of other studies. Several studies have demonstrated that milk thistle has protective properties against toxic agents. (Fanoudi et al., 2020). This study (Ali et al., 2014) reported that there is evidence that the milk thistle significantly improved elevated liver enzyme s alanine aminotransferase (ALT) and aspartate aminotransferase (AST). Also (Mumtaz et al., 2020) proved that garlic is recommended as an active antioxidant against heavy metal-induced toxicities. It might act as a lead chelator and enhance the excretion of lead and other heavy metals from the body. The natural chelating ability of allicin and sulfhydryl groups which are present in garlic makes it a strong antioxidant for the treatment of toxicity induced by lead, particularly prolonged toxicity of lead. Results indicate that

ginger has the potential to ameliorate lead induced hepatic injury due to oxidative stress in rats. Ginger may exert its protective actions against lead-induced hepatotoxicity in rats possibly through its antioxidant mechanisms and may have future therapeutic relevance (Atef et al., 2013). This study proved that the administration of Aloe. vera leaves aqueous extract was able to markedly protect rat liver from the lead-induced toxicity. (Gupta et al., 2019). In this study (Zhang et al., 2012) We found that the corn treatment prevented the elevation of serum aminotransferase and alleviated the hepatic histological damage that was induced by toxins. This study (Mahesh et al., 2016) proved that there was an improvement in liver function as indicated by decreased AST levels When eating carrots for 8 weeks. (Achuba et al., 2015) suggest that the consumption of natural honey supplemented diet has chemoprotective and ameliorative effects against toxicity on kidney and liver tissue damage. The liver is extremely sensitive to cadmium's toxic effects. This may be due to the ability of these tissues to synthesize metallothionein (MT), which are Cd-inducible proteins that protect the cell by tightly binding the toxic cadmium ions. (Genchi et al., 2020). Animal studies have shown that accumulation of Cd in the liver significantly increased the activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), lactate dehydrogenase and alkaline phosphatase. Various hepatic histopathological changes, including parenchymal damage and inflammatory cell infiltration, and activation of Kupfer cells. (Wang et al., 2021). Mercury chloride as one of the most toxic salts of mercury, is metabolized primarily in the liver and then, accumulated in the kidneys. Consequently, the liver and kidneys are considered the most affected organs (Nabil et al., 2020). The accumulation of mercury in the liver causes hepatotoxicity. (Hazelhoff et al., 2018) Exposure to a mixture of cadmium (Cd), lead (Pb) and mercury (Hg) can induce liver damage. (Zou et al., 2020). In the general, higher exposure to lead, cadmium, and mercury was positively

associated with biomarkers of liver injury including ALT and AST (Zhou et al., 2022).

Oxidative stress of rats fed on different combinations of foods.

The results of MAD activity and CAT enzyme of all tested groups are presented in Table (2). The antioxidant parameters, after 4 weeks post treatment with lead in the positive control group (5.07±0.56) there were statistically significant ($P \leq 0.05$) elevations in the activities of (MDA) when compared with negative control groups (0.75 ± 0.06) (which was fed the experimental diets). While there was a significant ($P \leq 0.05$) decrease in catalase activities in the positive control group (0.91±0.04) when compared with the negative control groups (4.35 ± 0.20) (which was fed the experimental diets). Results of MAD activity and CAT, after 4 weeks post-treatment with lead then 4 weeks of treatment with different combinations of plants in the treatment groups (2.47±0.06 and 1.96± 0.09) formula (1) and formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (MDA) when compared with positive control groups (5.07±0.56). While there were statistically significant ($P \leq 0.05$) elevations in the activities of catalase activities in the treatment groups (1.72±0.18 and 2.24±0.20) formula (1) and formula (2) when compared with positive control groups (0.91±0.04). There was a significant ($P \leq 0.05$) decrease in the activities of (MDA) While there were statistically significant ($P \leq 0.05$) elevations in the activities of catalase activities between formula (1) and formula (2) in lead fed groups in favor of formula two

Results of MAD activity and CAT enzyme, after 4 weeks post-treatment with cadmium in the positive control group (5.76±0.78) there were statistically significant ($P \leq 0.05$) elevations in the activities of (MDA) when compared with negative control groups (0.75 ± 0.06) (which was fed the experimental diets). While there was a significant ($P \leq 0.05$) decrease in catalase activities in the positive control group (0.78±0.04) when compared with the negative control groups (4.35 ± 0.20) (which was fed the experimental diets).

Catalase (CAT) enzyme activity as another main antioxidative element (Yaribeygi et al., 2018). Antioxidant enzymes have certain active sites which act as targets for toxic heavy metals like lead, mercury etc., resulting in a decrease in their enzymatic activities. Lead has a high affinity to sulfhydryl groups of these enzymes interrupting their functional properties (Amin et al., 2021). Lead can inhibit several antioxidant enzymes. The decrease in (CAT) activity after lead exposure might be due to an offset in the elimination of H2O2 and toxic peroxides which induce the accumulation of H2O2 by ROS. the inhibition of (SOD) activity increases the level of O2 resulting in an inactivation of CAT (Aouini et al., 2018). This is as shown in table (2), MDA activity and CAT enzyme, after 4 weeks post-treatment with cadmium then 4 weeks of treatment with different combination of plants in the treatment groups (3.64±0.10 and 2.29±0.11) formula (1) and formula (2) there were significant ($P \leq 0.05$) decrease in the activities of (MDA) when compared with positive control groups (5.07±0.56). While there were statistically significant ($P \leq 0.05$) elevations in the activities of catalase activities in the treatment groups (1.23±0.18 and 2.59±0.45) formula (1) and formula (2) when compared with positive control groups (0.78±0.04).

Lead chloride may induce oxidative stress leading to the generation of free radicals and alteration in oxygen free radical scavenging enzyme system or antioxidant and damage membrane structure (Sudjarwo and Sudjarwo, 2017). MDA is one of the major products of lipid peroxidation and can be used to evaluate membrane damage. Furthermore, changes in MDA content are typically used to indicate how an organism resists stress (Chen et al., 2015). The major pathway of Pb toxicity is to generate ROS, elevation lipid peroxidation (MDA), and depletion of the antioxidant system resulting in induction of oxidative stress organ damage (El-Boshy et al.,2019).

Table (2) Blood chemical analyses to clarify the effect of Lead poisoning on Oxidative stress Enzymes

Parameters	Oxidative stress (n. mol/mg/ protein)			
	Control (-)	Control (+)	Treated groups	
			Formula (1)	Formula (2)
Lead -fed groups				
MDA	0.75 ± 0.06 ^{d}	5.07±0.56^{a}	2.47±0.06^{b}	1.96± 0.09^{c}
CAT	4.35 ± 0.20 ^{a}	0.91±0.04^{d}	1.72±0.18^{c}	2.24±0.20^{b}
Cadmium -fed groups				
MDA	0.75 ± 0.06 ^{d}	5.76±0.78^{a}	3.64±0.10^{b}	2.29±0.11^{c}
CAT	4.35 ± 0.20 ^{a}	0.78±0.04^{d}	1.23±0.18^{c}	2.59±0.45^{b}
Mercury -fed groups				
MDA	0.75 ± 0.06 ^{d}	5.46±0.21^{a}	3.96±0.08^{b}	2.50±0.36^{c}
CAT	4.35 ± 0.20 ^{a}	0.82±0.08^{d}	2.00±0.15^{c}	2.99±0.25^{b}
Lead, Cadmium and Mercury -fed groups				
MDA	0.75 ± 0.06 ^{d}	6.40±0.33^{a}	4.58±0.43^{b}	3.13±0.39 ^{c}
CAT	4.35 ± 0.20 ^{a}	0.55±0.04^{d}	1.36±0.21^{c}	2.38±0.57 ^{b}

* Data are presented as means ± SDM.
MDA: Malondialdehyde CAT: Catalase

consistent with the findings of the current study. From these results we conclude that lead is postulated to be involved in the induction of oxidative stress within major organs of the body. (El-Boshy et al., 2019). Comparable results were obtained in a study by (Kasperczyk et al., 2015), who reported significantly increased MDA levels in erythrocytes from apprentices exposed to low-dose Pb increased MDA concentration. Animal studies have also reported similar findings. In rats that were orally exposed to lead acetate, lead nitrate, or lead chloride, many authors reported increased MDA levels in the liver (Wang et al., 2016).

In this study, lead was administered to rats at a dose of 50 mg/kg/day for six days which produced a significant elevation in blood lead level, a meaningful increase in MDA and a decrease in CAT compared to the control group (Mohammadi et al., 2022) In this study it has been reported that MDA level in renal tissues of CP-treated rats displayed a significant increase compared with

control. However, CAT activities revealed a significant reduction (Youssef et al., 2015). Garlic extract has beneficial effects against oxidative stress. (Hamza et al., 2017) Lines of evidence suggest that garlic extract (GE) is rich in antioxidants and able to scavenge ROS by amplification of intercellular antioxidant enzymes e.g., CAT (Ghalehkandi, 2018). This is consistent with many studies, one of which mentioned that the enzymes activity in the lead and garlic extract Groups increases significantly with decreased malondialdehyde (MDA) levels when compared with lead treated group only (Hamza et al., 2017). In the present study, a lead-induced significant increase was expressed in terms of a marked increase in MDA concentration However, ginger decrease MDA which may be due to the free radical scavenging property of ginger owing to its anti-oxidative nature (Amin et al., 2021). several previous studies demonstrated the anti-apoptotic activity of ginger. The oxidative stress was suppressed following combined treatment with ginger extract and garlic, as reflected by a significantly decreased MDA and markedly increased activity of antioxidants including CAT (Hemieda et al., 2019). Also, the milk thistle has an antioxidant effect and capacity to scavenge ROS. This finding agrees with other investigations, which showed that silymarin is a potent antioxidant and might act as an antigenotoxic agent. (Alcaraz-Contreras et al., 2016). This study reported that the treatment of rats with carrots at a dose of 400 mg/kg BW prevented the levels of MDA to rise when the rats were challenged with lead acetate. This means that carrots minimized the toxic effect of lead acetate via its antioxidant activity. (Sudjarwo and Sudjarwo, 2017). The rise in MDA levels in the lead group could be a sign of OS damage. However, treatment with honey prevent MDA levels from rising. (Adeyomoye et al., 2022). Oxidative stress is another manifestation of Cadmium damage and has been found to play an important role in the toxicity of many xenobiotics. The present investigation showed a significant increase in levels of malondialdehyde (MDA) (Amamou et al., 2015). This is consistent with the findings of the current study. The results of

this study agree with (Unsal et al., 2015) who stated that cadmium exposure significantly increased the tissue MDA levels ($p < 0.01$) and significantly decreased the tissue CAT activities ($p < 0.01$) when compared with controls after Rats intoxicated with Cadmium for 30 days, significantly increased tissue malondialdehyde (MDA) levels and significantly decreased enzymatic antioxidants catalase. Like our findings (Kassab et al., 2020) confirm the ability of Cd to induce oxidant/antioxidant imbalance, witnessed by increased MDA lowered CAT level antioxidant enzymatic activities.

Histopathological examination

The liver was examined by a histological approach and the photomicrographs of hematoxylin – eosin-stained liver is illustrated.

Liver

The liver sections from the control positive group (normal rats fed on a commercial diet only) showed the normal histological structure of the central vein and surrounding hepatocytes in the hepatic parenchyma (Fig 1). And we note in the control (+) group of rats induced by Lead dilatation and congestion were detected in the central vein, hepatic sinusoids, and portal vein with dilatation of the bile ducts. Fine fibroblastic cells proliferation was detected in the portal area (Fig 2). While, in control (+) groups of rats induced by cadmium has been noticed severe congestion and dilatation were detected in the central veins associated with diffuse degeneration in the hepatocytes all over the parenchyma. The portal area showed multiple numbers of newly formed bile ductulus and congestion in the portal vein (Fig 6). Also, the control (+) group of rats induced by mercury showed severe dilatation and congestion detected in the central and portal vein, associated with degeneration in a diffuse manner all over the hepatocytes in the parenchyma. The portal area showed hyperplasia in the bile ducts with periductal inflammatory cells infiltration and oedema in the

portal area (Fig 10). As well, in the control (+) group of rats induced by lead, cadmium, and mercury saw the central veins and sinusoids showed dilatation and congestion, associated with focal necrosis in the hepatocytes in the parenchyma. The portal area showed periductal oedema and inflammatory cells infiltration surrounding the bile ducts as well as dilatation in the portal vein. Few fibroblastic cells proliferation was originated from the portal area and extended in between the hepatocytes in the parenchyma (Fig 14).

On the other hand, in a group of rats experimentally induced with lead and then treated by formula (1) it was noticed severe dilatation was detected in the central vein. while the portal area showed thickening and fibrosis with collagen proliferation as well as congestion in the portal vein (Fig 3). Likewise, a group of rats experimentally induced with lead and then treated by formula (2) showed no histopathological alteration (Fig 4). Ditto in the group of rats experimentally induced by cadmium then treated by formula (1) it was noticed Dilatation was detected in the central vein, while the hepatocytes in the parenchyma was intact (Fig 7). Also, a group of rats experimentally induced with cadmium and then treated by formula (2) showed no histopathological alteration. (Fig 8) Moreover, a group of rats experimentally induced with mercury and then treated by formula (1) it was noticed, the portal area showed congestion in the portal vein and hyperplasia in the bile ducts (Fig 11). Also, a group of rats experimentally induced by mercury then treated by formula (2) showed no histopathological alteration. (Fig 12). Whereas, in group of rats experimentally induced by lead, cadmium and mercury then treated by formula (1) it was noticed, no histopathological alteration (Fig 15). The same result, in group of rats experimentally induced by lead, cadmium and mercury then treated by formula (2) showed no histopathological alteration. (Fig 16). Cadmium accumulation in liver is a well-documented event and considered to be an important mechanism of hepatic damage induced by this metal (El-Sokkary et al., 2010) This study

reported that histologic analysis showed that the control group had normal architecture, whereas the introduction of Cd and Hg induced pathologic changes in liver (Raghuvanshi et al., 2016). Cadmium and lead caused damage to liver cells in rats by activating the body's antioxidant system and inducing autophagy. The combined treatment of Cd with Pb exacerbated this change. (Zou et al., 2020). There is evidence that chronic exposure to low concentrations of mercury causes tissue or liver damage (Kumar et al., 2014). Results revealed that garlic ameliorated liver damage induced by HgCl2 (El-Shenawy et al., 2008). Also ginger offered more protective ability through the elimination of Cd and Hg, it significantly reduced the accumulation of Pb in the liver. (Nwokocha et al., 2012). This study provided a considerable support for evidencing the protective effects of silymarin on liver damage induced by heavy metals. (Chtourou et al., 2013). The reduction in some of the damage caused by heavy metals administration may be attributed to the beneficial effect of honey and aloe vera in the prevention of hepatic damage induced by obstruction of the common bile duct results. (Garba et al., 2012)

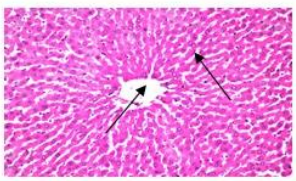

Fig. 1. Control (-ev) there was no histopathological alteration and the normal histological structure of the central vein.

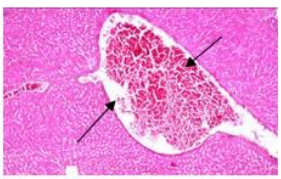

Fig. 2. Control (+ve) induced by lead showed dilatation and congestion were detected in the central vein.

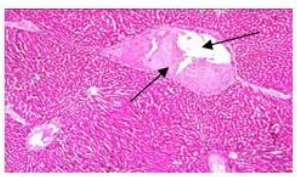

Fig. 3 Treated by formula (1) showed sever dilatation was detected in the central vein.

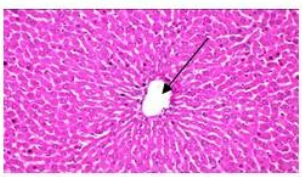

Fig. 4 Treated by formula (2) showed no histopathological alteration.

Histological changes in the liver stained with (Hand E X40) using different formulas of food in lead fed groups.

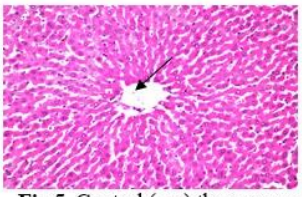

Fig.5. Control (-ev) there was no histopathological alteration and the normal histological structure of the central vein.

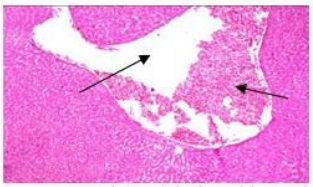

Fig. 6. Control (+ev) induced by cadmium sever congestion and dilatation were detected in the central veins.

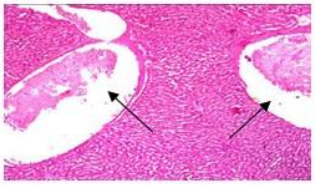

Fig. 7. Treated by formula (1) showed dilatation was detected in the central vein

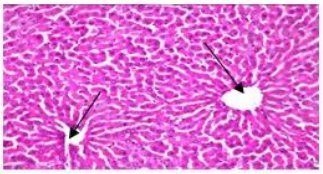

Fig. 8 Treated by formula (1) showed no histopathological alteration

Histological changes in the liver stained with (Hand E X40) using different formulas of food in cadmium fed groups.

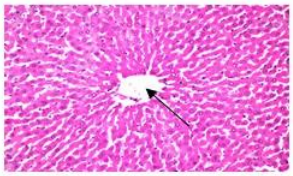

Fig. 9 Control (-ev) there was no histopathological alteration and the normal histological structure of the central vein and surrounding hepatocytes in the parenchyma.

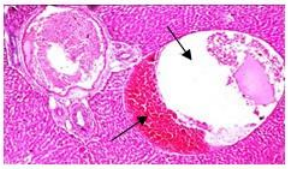

Fig. 10. Control (+ev) induced by mercury showed sever dilatation and congestion were detected in the central and portal vein.

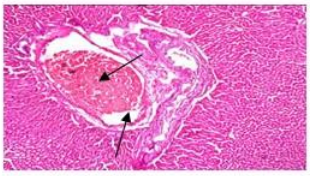

Fig.11. Treated by formula (1) showed sever dilatation was detected in the central vein.

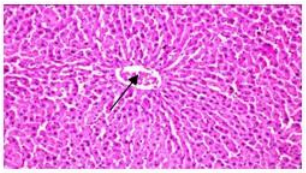

Fig. 12. Treated by formula (2) showed no histopathological alteration.

Histological changes in the liver stained with (Hand E X40) using different formulas of food in mercury fed groups

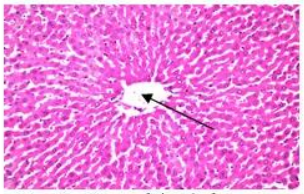

Fig.13. Control (-ev) there was no histopathological alteration and the normal histological structure of the central vein and surrounding hepatocytes in the parenchyma.

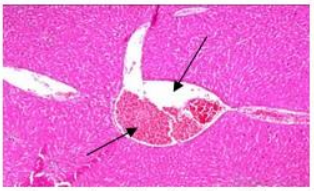

Fig. 14. Control (+ev) induced by lead, cadmium, and mercury showed central veins and sinusoid showed dilatation and congestion.

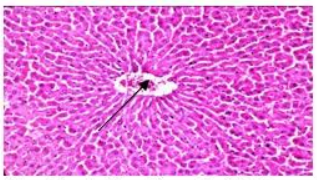

Fig. 15. Treated by formula (1) showed no histopathological alteration.

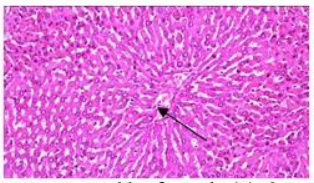

Fig. 16. Treated by formula (1) showed no histopathological alteration.

Histological changes in the liver stained with (Hand E X40) using different formulas of food in lead, cadmium, and mercury fed groups.

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