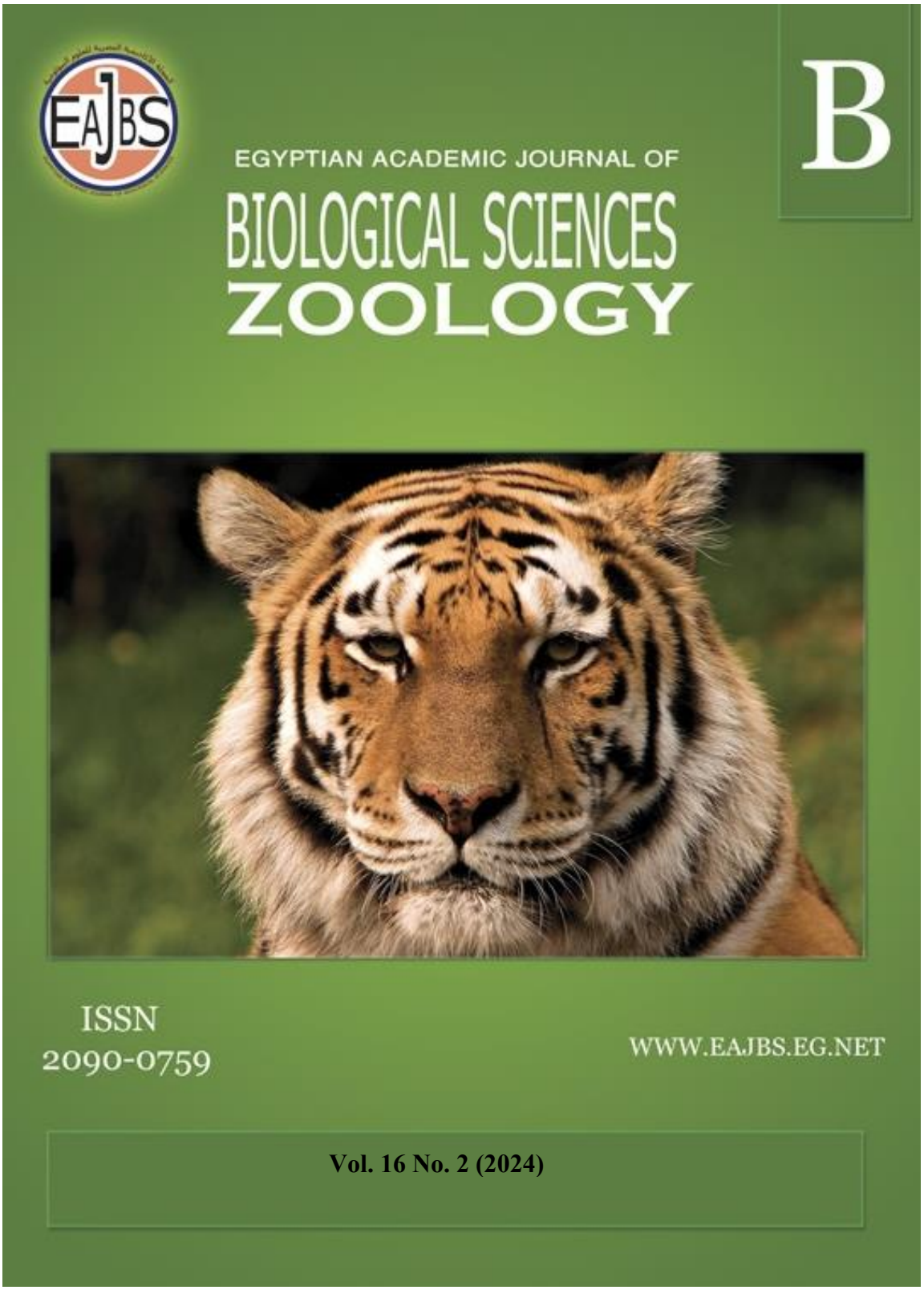

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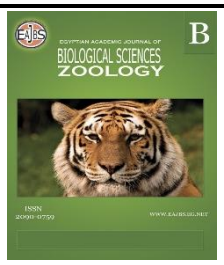

The Protective Effect of Soybean (*Glycine max*) Oil on Male Rats Exposed to Cadmium

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ABSTRACT

Background: Cadmium (Cd^{+2}) is a substantial environmental contaminant acknowledged for its extensive adverse effects. *Glycine max (GM*), native to Asia, is recognized for its hepatoprotective and antioxidative properties. **Objective**: The current study investigates the protective influences of *Glycine max* on cadmium (Cd^{+2})-triggered oxidative stress in the hepatic tissue of male rats. **Methods:** Cd^{+2} (5 mg/kg) was given orally for six weeks to induce hepatotoxicity, followed by *Glycine max* oil (300 mg/kg) treatment. **Results:** The results indicated that cadmium caused damage to the hepatic cells, leading to an increase in the levels of aspartate transaminase (AST), alanine transaminase (ALT), total protein (T.P), and serum total bilirubin (T. Bil), and a decrease in albumin. The detrimental effect of Cd^{+2} was evident in the substantially reduced levels of glutathione (GSH) and superoxide dismutase (SOD). The activity of hepatic enzymes was restored to near-normal levels when cadmium-induced rats were administered with 300 mg/kg *Glycine max* oil. Additionally, *Glycine max* oil significantly restored the antioxidant levels in the hepatic tissue. Histopathological examinations of rat livers demonstrated that *Glycine max* (300 mg/kg) significantly mitigated the influence of Cd^{+2} toxicity and preserved the tissue's typical histological structure. **Conclusion:** Recent research has suggested that *Glycine max* potentially reduces the oxidative damage caused by cadmium in rats' hepatic tissue.

INTRODUCTION

A substantial source of health issues for individuals has been the emergence of environmental toxicants, adversely impacting the health and overall well-being of people. Recently, industrial activity has increased, resulting in extended human exposure to industrial pollutants, including Cd^{+2}, that is utilized in the manufacture of batteries, the creation of pigments, electroplating applications, refineries, plastics, and petrochemical fields, and

cigarette smoke (Genchi *et al.,* 2020a). Exposure to Cd^{+2} has been documented in different forms throughout the last century, with Cd^{+2} prevalent in the environments associated with various activities carried out by humans. Cd^{+2} is a persistent heavy metal with a long half-life, accumulating in biological tissues and causing toxicity and dysfunction. Cd^{+2} exhibits many toxicological pathways in certain species within controlled experimental circumstances, and it is a prevalent hazardous element that induces the generation of free radicals and increases reactive oxygen species (ROS), including hydrogen peroxide and thiobarbituric acid, which compromise the antioxidant defense system (Bull & Chapd, 2010; Renugadevi & Prabu, 2010).

Moreover, Cd^{+2} absorption in plant and animal tissues constitutes an additional pathway for human exposure to this contaminant. As a result of the genetic and epigenetic influence of Cd^{+2}-triggered toxicity, biological indicators are being studied for low levels of cadmium as a water contaminant (Khalil *et al.,* 2014). Prolonged exposure to cadmium by humans may harm several organs, including the lungs, gastrointestinal tract, nervous system, testes, immunological system, endocrine system, kidneys, and liver (Unsal *et al.,* 2020). Moreover, Cd^{+2} is a potential carcinogen that induces oxidative damage in the blood and other organs, resulting in cellular membrane malfunction (Jemai *et al.,* 2007). Recent studies demonstrate that Cd^{+2} may produce several variations in epigenetic and genetic in both plant and mammalian cells, in both *vivo* and in *vitro* (Genchi *et al.,* 2020b).

Urine enzymes (lactate dehydrogenase, alkaline phosphatase, and N-acetyl-beta-D-glucosaminidase) rise following a 10-month exposure to Cd^{+2} (Groten *et al.,* 1994). Chronic exposure to Cd^{+2} in hepatic tissue has resulted in elevated lipid peroxidation and alterations in critical components. Furthermore, Cd^{+2} may disrupt hepatic cellular respiration (Müller & Ohnesorge, 1984). Consequently, many pathological hepatocyte alterations have been seen because of Cd^{+2} poisoning, featuring granulomatous inflammation, cellular proliferation, nodular hyperplasia, programmed cell death, and necrosis. Morphological alterations occur with elevated blood levels of liver enzymes, such as ALT and AST (Jeong *et al.,* 2000).

Glycine max is a plant that originated in East Asia and is extensively farmed for its edible beans. It is preferred by a broad range of temperatures and soils, hence it is regarded as the cheapest crop (Zaefarian & Rezvani, 2016). It comprises around 20 % oil and 40 % vital proteins. *Glycine max* protein comprises high critical amino acids (5%), whereas most cereals are insufficient. Furthermore, it includes many minerals, unsaturated fats, salts, and vitamins (thiamine, riboflavin), and its germinated grains contain a sufficient amount of vitamin C (Miransari, 2016; Imtiyaz *et al.,* 2014). This study examines the impact of *Glycine max* oil treatment on mitigating chronic liver toxicity caused by Cd^{+2} and enhancing hepatic tissue and oxidant/antioxidant status rates.

MATERIALS AND METHODS

Animals*:*

The rats were obtained from the Animal Unit of the King Fahd Medical Research Center at King Abdulaziz University in Jeddah, Saudi Arabia. Male albino rats (130–150 g) were chosen for the investigation and kept within appropriate stainless-steel cages within a conventional laboratory environment (maintained at 25 ± 2°C, with 50 ± 5% relative humidity, and a 12-hour light/dark cycle). The rats were fed with ordinary rat chow and allowed to drink tap water anytime. Rats were accustomed to lab atmospheres for 7 days. The experiments were processed using the animal ethical rules of King Abdulaziz University's Animal Care and Use Committee (ACUC). Furthermore, all tests adhered to the Arrive standards and the EU Directive 2010/63/EU regarding animal research.

Experimental Design:

Albino rats were categorized into four groups, including ten rats per group. The

subsequent doses of *Glycine max* oil were delivered daily for six consecutive weeks as follows:

Group I: Control group; oral (0.9% NaCl).

Group II: Cd^{+2} group; oral Cd^{+2} (5 mg/kg daily).

Group III: Cd^{+2} + *Glycine max* group; oral Cd^{+2} (5 mg/kg daily) + oral *Glycine max* oil (300 mg/kg daily).

Group IV: *Glycine max* group; oral saline (0.9% NaCl) + oral *Glycine max* oil (300 mg/kg daily).

Liver Function:

The levels of aspartate transaminase (AST) and alanine transaminase (ALT)were tested in serum using the modified kinetic method (Tietz, 1976). Albumin was measured utilizing Biocon Company's kits using dye-binding procedures for bromocresol purple (BCP), as reported by (Carter, 1970; Louderback & Shanbrom, 1968); total proteins (TP) were evaluated according to the technique described by Marshall & Williams, (2003). Total bilirubin was detected in blood serum using the method specified by Bonora *et al.* (1983).

Antioxidant Parameters:

The antioxidant markers superoxide dismutase (SOD) and glutathione (GSH) were selected to evaluate oxidative stress. SOD activity was measured using the methods of Misra & Fridovich (1972) and Greenwald (1985). GSH levels were assessed using the protocols described by Moron *et al.* (1979) and Wendel (1981). *Liver histopathological examination*

Liver tissues were fixed in a 10% formalin solution, dehydrated in various alcohol concentrations, cleaned with xylol, and embedded in paraffin wax. The histological investigation used 5 µM sections stained with Harris haematoxylin and eosin.

Statistical Analysis:

Data is presented as mean ± standard deviation (SD) for comparison. The findings were analysed statistically using Prism® software for Windows, version 9.50 (GraphPad, USA). Results were assessed for significance with $P < 0.05$ using one-way ANOVA.

RESULTS

Liver Function:

Figure 1 (A-E), depicts control and experimental rats' blood hepatic marker enzyme levels, albumin, and total bilirubin. Cd^{+2} treatment significantly impaired liver function in experimental rats. Cd^{+2}-intoxicated animals had substantially higher levels of ALT, AST and total bilirubin (T.B) versus the control group ($P < 0.05$). Administration of *Glycine max* oil (300 mg/kg) oil to Cd^{+2}-induced rats significantly reduced ($P < 0.05$) blood liver enzymes and T.B while increasing albumin levels compared to rats exposed to Cd^{+2}. The dosage (300 mg/kg) restored ALT, AST, albumin, and total bilirubin to normal levels compared to Cd^{+2} induced rats. As a result of these results, 300 mg/kg was chosen as the dosage for the next tissue study (Fig. 2).

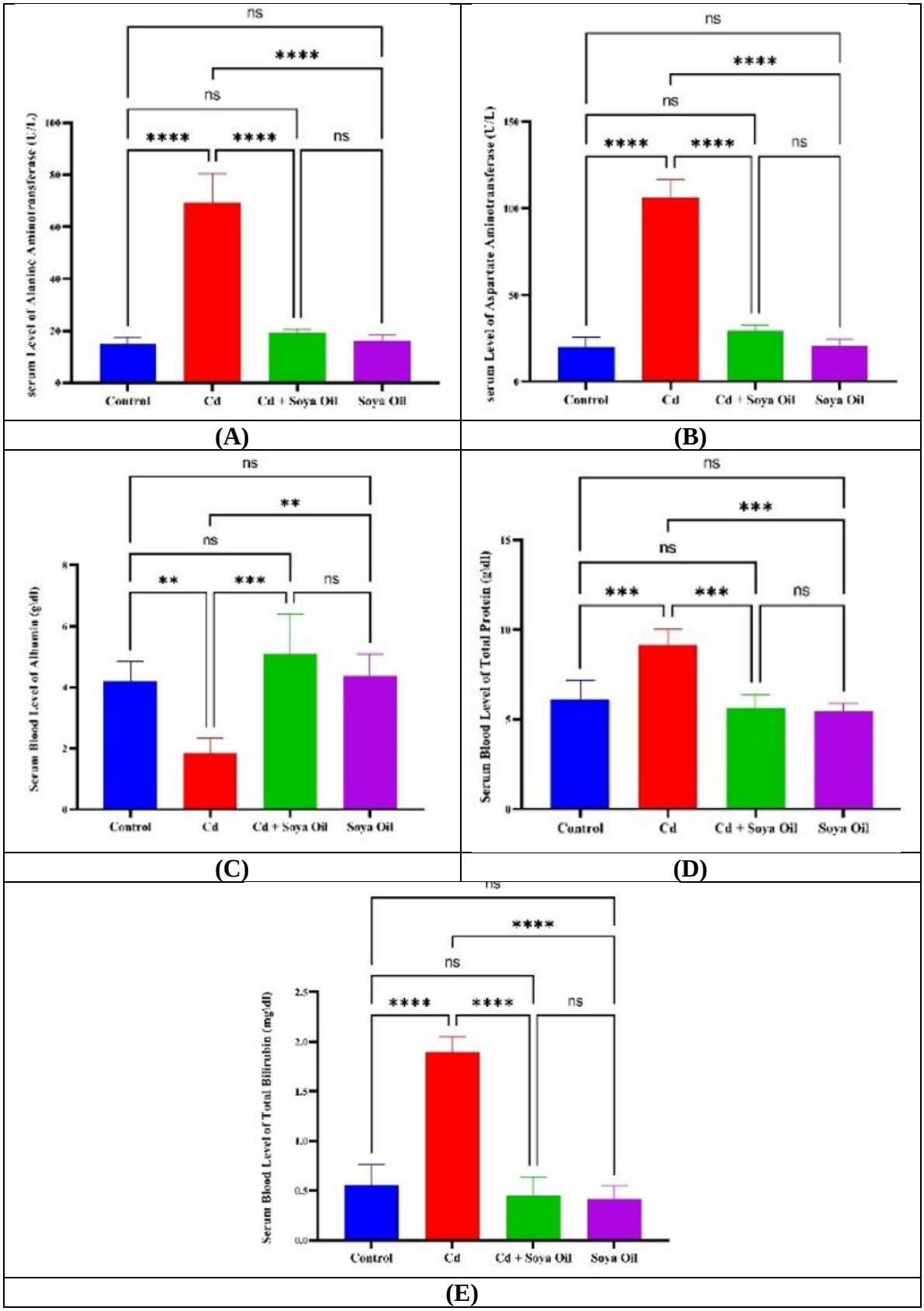

Fig. 1. Serum levels of liver function A) represent ALT, B) represent serum AST, D) represent serum T.P, and E) represent serum T.B in different groups being studied. Findings are presented as mean +/- standard deviation. A significance was conducted at $P < 0.05$.

Hepatic Antioxidant Measurement:

Figure 2. (A, B), illustrates the change in GSH and SOD levels in the Cd+2-induced rats' livers. A significant depletion ($P < 0.05$) in the GSH and SOD levels was detected in rats exposed to Cd^{+2} compared to normal untreated rats. Treatment via *Glycine max* oil (300 mg/kg) significantly restored GSH and SOD levels to near-normal values when compared to untreated rats.

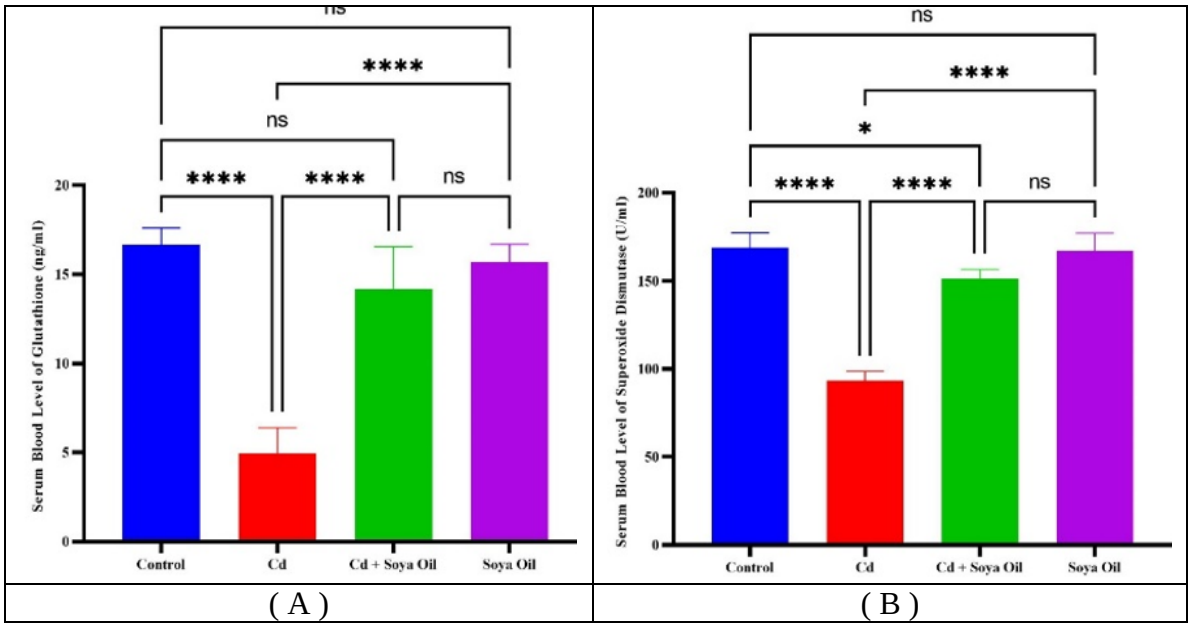

Fig. 2. Blood serum measurements of oxidative stress markers A) represent serum GSH and B) represent SOD in different groups being studied. Findings are presented as mean +/- standard deviation. A significance was conducted at $P < 0.05$.

Histopathology:

Histopathological examinations demonstrated that exposure to Cd^{+2} revealed liver injury, including sinusoidal dilation, fatty degeneration, and necrosis (Figs. 3B, C) versus untreated control liver (Fig. 3 A). The injuries were decreased in the hepatic tissue of rats treated with *Glycine max* and induced with Cd^{+2} (Fig. 3 C). The histological examination was roughly normal in rats treated with *Glycine max* alone (Fig. 3 D).

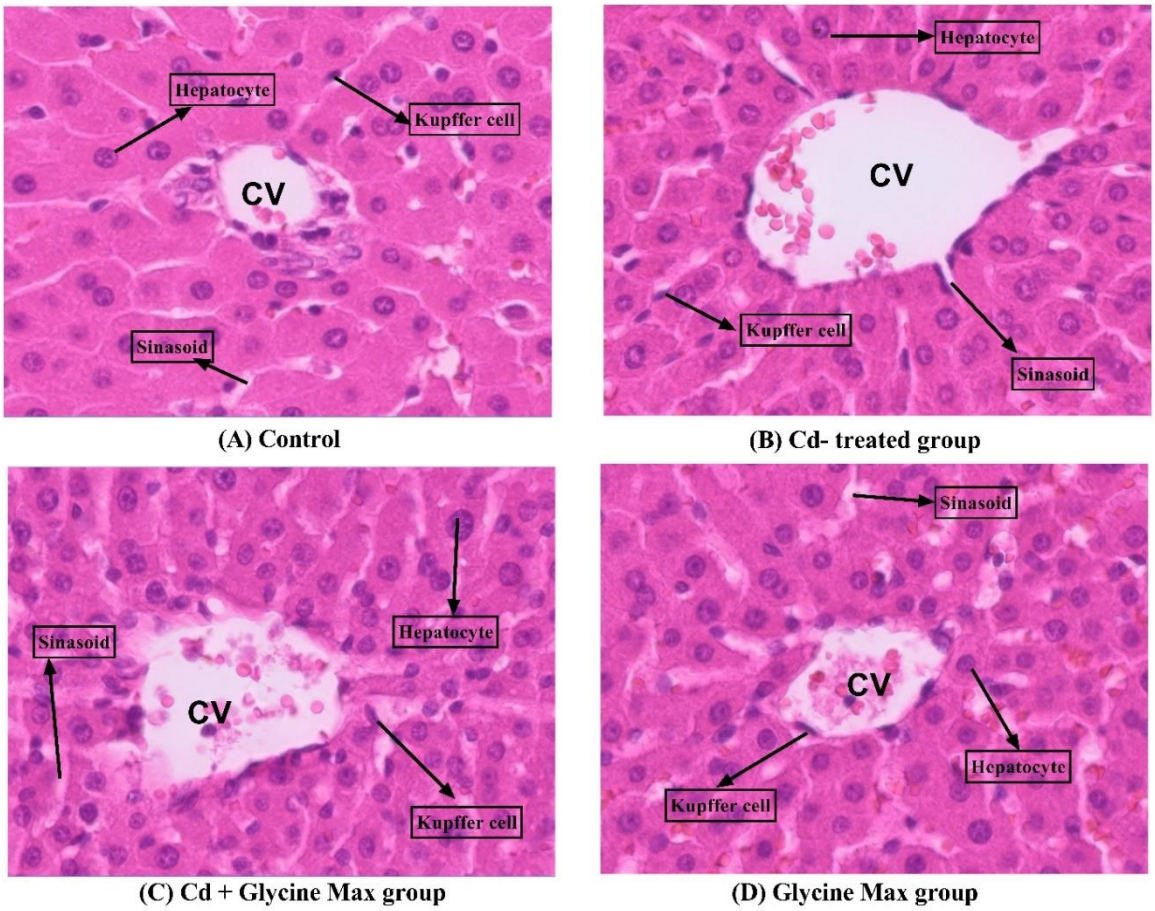

Fig. 3. Photographs representing the hepatic tissue demonstrate *Glycine max* oil's potential effect on Cd^{+2} -induced Rats. A) Untreated control liver (40 x). B) Cd^{+2} exposure in rat liver causes a noticeable alteration in the histological pattern of hepatic tissue. C) Treatment with *Glycine max* rats induced with Cd^{+2} improved liver tissue effectively. D) The examination of the untreated group and the group treated with *Glycine max* oil groups looked similar, and no noticeable change was detected. Cd^{+2}: Cadmium, CV: central vein.

DISCUSSION

Cadmium, a hazardous metal, is extensively utilized in various sectors. It increases early oxidative stress and then leads to the progression of severe pathological diseases due to its lengthy accumulation in specific tissues (Bagchi *et al.*, 2000). The liver is the vital organ for the metabolism of xenobiotics and the essential target site that reduces the toxicity of Cd^{+2} (Liu *et al*., 2009). Chronic exposure to Cd^{+2} may cause nephrotoxicity, hepatotoxicity, damage to the neurological system, immunological and endocrine systems, and, subsequently, cancer in humans (Hartwig, 2013; Newairy *et al.*, 2007). Several mechanisms involving an increase in lipid peroxidation and an interaction with membrane compositions have been suggested to explain Cd^{+2}-induced liver damage.

It is claimed that Cd^{+2} triggers oxidative stress and lipid peroxidation *via* decreasing GSH or suppressing antioxidant enzymes (Bagchi *et al.*, 1996). Previous studies have demonstrated that antioxidants can lessen the damaging effects of Cd^{+2} exposure by reducing the metabolic changes in the body. Therefore, antioxidant supplementation could deliver protection against cadmium poisoning (Karbownik *et al.*, 2001). *Glycine max* provides essential minerals, including magnesium, phosphorus, iron, calcium, manganese, and phosphate, with a high potassium concentration. Additionally, it supplies vitamins B6 and E

(Caicedo *et al.,* 2019; Tosquy-Valle *et al.,* 2010). Several studies have investigated soy's bioactive components, which have bioactive components, including proteins, fats, fiber, and phytochemicals like isoflavones. Soybeans are rich in three main isoflavones: genistein, daidzein, and glycitein. Notably, it has been suggested to act as an inhibitor of oxidative stress, angiogenesis and metastasis, (Mezei *et al.,* 2003). In this respect, the current research also found that administering *Glycine max* oil (300 mg/kg) significantly protected hepatic function from the harmful effects of Cd^{+2}.

Elevated serum hepatic marker enzyme levels, which show cellular leakage and loss of functional integrity of the hepatic membrane architecture, indicate liver injury following Cd^{+2} exposure. Elevated ALT and AST levels are essential for identifying liver injury (Williamson *et al.,* 1996). Renuka *et al.* (2021) reported that total bilirubin and liver function enzymes (ALT, AST) were significantly elevated during Cd^{+2} poisoning. In contrast, albumin and other proteins were significantly decreased (Lovásová *et al.,* 2013). There was an agreement that hepatic function enzymes are trustworthy indicators of liver health. In the current investigation, the blood level of total bilirubin was also shown to increase in Cd^{+2} -intoxicated rats. This is consistent with the results of Liss *et al.* (1985), who proposed that elevated serum T.B. is a clear indicator of liver dysfunction. Cd^{+2} poisoning has been shown to drastically increase blood hepatic marker enzymes and total bilirubin levels (Prabu *et al.,* 2008). Increased concentrations of these hepatic enzymes signify the pathological condition of hepatic tissues and the consequent hepatic impairment (Kandemir *et al*., 2020).This aligns with the findings of the current investigation, which found elevated AST, ALT, T. P, and T. Bil activity, as well as lower Alb levels in the serum of Cd^{+2}-treated rats.

The excessive production of reactive oxygen species (ROS) impairs the functionality of cellular organelles, such as mitochondria and the endoplasmic reticulum, as well as DNA, thus influencing protein synthesis. These activities cause necrotic damage to liver tissues and impair hepatic functioning, leading to decreased albumin levels and total proteins (Dwivedi *et al.,* 2015; Ojo *et al.,* 2014). The current study revealed that Cd^{+2} exposure markedly reduced SOD activity and GSH levels, signifying oxidative stress. SOD is regarded as an antioxidant; it is classified as a primary endogenous antioxidant enzyme that possesses a robust ability to combat ROS (Ighodaro & Akinloye, 2018). Superoxide ions are produced as a secondary product during oxidative stress and convert into H_2O_2 (Kheradmand *et al.*, 2010). GSH is present in each biological system. By directly engaging its sulfhydryl group with ROS, it functions as a non-enzymatic antioxidant and is the primary defense against oxidative injury. It may serve as a cofactor or coenzyme in the enzymatic detoxification of ROS. Cadmium is predominantly bound to the sulfhydryl groups of GSH, which is why it is inactive (Sunitha *et al.,* 2001). In this study, the liver's susceptibility to free radical injury may be exacerbated by the reduced levels of GSH in Cd^{+2} toxicity. Our findings are consistent with earlier published research, which reveals that GSH concentrations decrease following Cd^{+2} poisoning (Pari & Murugavel, 2005). However, administering *Glycine max* oil (300 mg/kg) boosted the activity of SOD and GSH. Nonetheless, administering *Glycine max* to rats substantially reduced the hepatic marker levels. *Glycine max* has been shown to repair the structural integrity of hepatocyte plasma membranes by decreasing ROS levels, which may ultimately diminish the elevation of ALT, AST, and ALP into the bloodstream. Moreover, the T.B concentration diminished, while the albumin concentration augmented due to the ROS scavenging properties of *Glycine max.*

This study confirms that *Glycine max* oil effectively counters Cd^{+2}-induced liver damage by enhancing antioxidant defenses and mitigating oxidative stress. These findings provide new insights into the therapeutic potential of *Glycine max* as a protective agent against heavy metal toxicity. Future research could explore its efficacy in other organ systems affected by Cd^{+2} and investigate the molecular pathways underlying its antioxidant properties.

Conclusion

Overall, Cd^{+2} exposure caused hepatotoxicity in rats by increasing liver function enzymes and total bilirubin levels. In contrast, albumin levels and SOD and GSH activities were lowered. Cd^{+2} exposure induced histological damage in the rat liver. However, *Glycine max* oil administration significantly reversed all Cd^{+2}-induced liver damage in rats, owing to its antioxidant hepatoprotective properties. Finally, treatment with *Glycine max* oil is suggested as a promising therapeutic method for reducing Cd^{+2}-induced liver damage.

Declarations:

Ethical Approval: The experiments were processed using the animal ethical rules of King Abdulaziz University's Animal Care and Use Committee (ACUC). Furthermore, all tests adhered to the Arrive standards and the EU Directive 2010/63/EU regarding animal research.

Competing interests: The authors declare there are no conflicts of interest regarding the publication of this article.

Author's Contributions: The authors have equal distributions.

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References

Bagchi, D., Bagchi, M., Hassoun, E., & Stohs, S. (1996). Cadmium-induced excretion of urinary lipid metabolites, DNA damage, glutathione depletion, and hepatic lipid peroxidation in Sprague-Dawley rats. *Biological Trace Element Research, 52*, pp. 143-154.

Bagchi, D., Bagchi, M., Stohs, S. J., Das, D. K., Ray, S. D., Kuszynski, C. A., Joshi, S. S., & Pruess, H. G. (2000). Free radicals and grape seed proanthocyanidin extract: importance in human health and disease prevention. *Toxicology*, *148*(2-3), pp. 187-197.

Bonora, R., Malchiodi, A., & Panteghini, M. (1983). A single reagent method for determination of total and conjugated bilirubin with a centrifugal analyzer. *Clinica Chimica Acta*, 128(2-3), pp. 407-414.

Bull, S., & Chapd, H. (2010). Cadmium, toxicological overview. *Journal Health Protection Agency, 2*(1), pp. 1-15.

Caicedo, I. M., Gonzales, A. M., & Castro, F. B. (2019). Industrialización del cultivo de soya. *Observatorio de la Economía Latinoamericana* (11), 26.

Carter, P. (1970). Ultramicroestimation of human serum albumin: binding of the cationic dye, 5, 5′-dibromo-o-cresolsulfonphthalein. *Microchemical Journal*, *15*(4), pp. 531-539.

Dwivedi, V., Mishra, J., & Shrivastava, A. (2015). Efficacy study of livartho against paracetamol induced hepatotoxicity in adult Sprague Dawley rats. *Journal of Drug Metabolism & Toxicology*, *5*(6), pp. 1-7.

Genchi, G., Carocci, A., Lauria, G., Sinicropi, M. S., & Catalano, A. (2020a). Nickel: Human health and environmental toxicology. *International Journal of Environmental Research and Public Health*, 17(3), pp. 679.

Genchi, G., Sinicropi, M. S., Lauria, G., Carocci, A., & Catalano, A. (2020b). The effects of cadmium toxicity. *International Journal of Environmental Research and Public Health*, 17(11), 3782.

Greenwald, R.A. (1985). Handbook Methods for Oxygen Radical Research (1st ed.). *CRC Press*. https://doi.org/10.1201/9781351072922

Groten, J., Koeman, J., Van Nesselrooij, J., Luten, J., van Vlissingen, J. F., Stenhuis, W., & Van Bladeren, P. (1994). Comparison of renal toxicity after long-term oral

administration of cadmium chloride and cadmium-metallothionein in rats. *Toxicological Sciences*, 23(4), pp. 544-552.

Hartwig, A. (2013). Cadmium and cancer. Cadmium: from *Toxicity to Essentiality*, 11, 491-507.

Ighodaro, O., & Akinloye, O. (2018). First-line defense antioxidants-superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPX): Their fundamental role in the entire antioxidant defense grid. *Alexandria Journal of Medicine*, 54(4), pp. 287-293.

Imtiyaz, S., Agnihotri, R. K., Ganie, S. A., & Sharma, R. (2014). Biochemical response of Glycine max (L.) Merr. To cobalt and lead stress. *Journal of Stress Physiology & Biochemistry, 10* (3), pp. 259-272.

Jemai, H., Messaoudi, I., Chaouch, A., & Kerkeni, A. (2007). Protective effect of zinc supplementation on blood antioxidant defense system in rats exposed to cadmium. *Journal of Trace Elements in Medicine and Biology*, 21(4), pp. 269-273.

Jeong, S.-H., Habeebu, S. S., & Klaassen, C. D. (2000). Cadmium decreases gap junctional intercellular communication in mouse liver. *Toxicological Sciences*, 57(1), pp. 156-166.

Kandemir, F. M., Yıldırım, S., Kucukler, S., Caglayan, C., Darendelioğlu, E., & Dortbudak, M. B. (2020). Protective effects of morin against acrylamide-induced hepatotoxicity and nephrotoxicity: A multi-biomarker approach. *Food and Chemical Toxicology*, 138, 111190.

Karbownik, M., Gitto, E., Lewinski, A., & Reiter, R. J. (2001). Induction of lipid peroxidation in hamster organs by the carcinogen cadmium: amelioration by melatonin. *Cell Biology and Toxicology*, 17, 33-40.

Khalil, M., Furness, D. N., Zholobenko, V., & Hoole, D. (2014). Effect of tapeworm parasitisation on cadmium toxicity in the bioindicator copepod, Cyclops strenuus. *Ecological Indicators*, 37, pp. 21-26.

Kheradmand, A., Alirezaei, M., & Birjandi, M. (2010). Ghrelin promotes antioxidant enzyme activity and reduces lipid peroxidation in the rat ovary. *Regulatory Peptides, 162*(1-3), pp. 84-89.

Liss, G. M., Greenberg, R. A., & Tamburro, C. H. (1985). Use of serum bile acids in the identification of vinyl chloride hepatotoxicity. *The American Journal of Medicine*, 78(1), pp. 68-76.

Liu, J., Qu, W., & Kadiiska, M. B. (2009). Role of oxidative stress in cadmium toxicity and carcinogenesis. *Toxicology and Applied Pharmacology*, 238(3), pp. 209-214.

Louderback, A. L., & Shanbrom, E. (1968). Lactic dehydrogenase isoenzyme electrophoresis. *Journal of the American Medical Association*, 205(5), pp. 294-295.

Lovásová, E., Rácz, O., Cimboláková, I., Nováková, J., Dombrovský, P., & Ništiar, F. (2013). Effects of chronic low-dose cadmium exposure on selected biochemical and antioxidant parameters in rats. *Journal of Toxicology and Environmental Health*, Part A, 76(17), pp. 1033-1038.

Marshall, T., & Williams, K. M. (2003). Aminoglycoside interference in the pyrogallol red-molybdate protein assay is increased by the addition of sodium dodecyl sulfate to the dye reagent. *Clinical Chemistry*, 49(12), pp. 2111–2112.

Mezei, O., Shay, N., Banz, W. J., Steger, R. W., Peluso, M. R., & Winters, T. A. (2003). Soy isoflavones exert antidiabetic and hypolipidemic effects through the PPAR pathways in obese Zucker rats and murine RAW 264.7 cells. *The Journal of Nutrition*, 133(5), pp. 1238-1243.

Miransari, M. (2016). Soybean, protein, and oil production under stress. In Environmental Stresses in Soybean Production, 2, pp. 157-176. *Elsevier*.

Misra, H. P., & Fridovich, I. (1972). The role of superoxide anion in the autoxidation of

epinephrine and a simple assay for superoxide dismutase. *Journal of Biological Chemistry*, 247(10), pp. 3170-3175.

Moron, M. S., Depierre, J. W., & Mannervik, B. (1979). Levels of glutathione, glutathione reductase and glutathione S-transferase activities in rat lung and liver. *Biochimica et Biophysica Acta (BBA)-General Subjects*, 582(1), pp. 67-78.

Müller, L., & Ohnesorge, F. K. (1984). Cadmium-induced alteration of the energy level in isolated hepatocytes. *Toxicology*, 31(3-4), pp. 297-306.

Newairy, A., El-Sharaky, A., Badreldeen, M., Eweda, S., & Sheweita, S. (2007). The hepatoprotective effects of selenium against cadmium toxicity in rats. *Toxicology*, 242(1-3), pp. 23-30.

Ojo, O. A., Ajiboye, B. O., Oyinloye, B. E., Ojo, A. B., & Olarewaju, O. I. (2014). Protective effect of Irvingia gabonensis stems bark extract on cadmium-induced nephrotoxicity in rats. Interdisciplinary *Toxicology*, 7(4), pp. 208-214.

Pari, L., & Murugavel, P. (2005). Role of diallyl tetrasulfide in ameliorating the cadmium-induced biochemical changes in rats. *Environmental Toxicology and Pharmacology*, *20*(3), pp. 493-500.

Prabu, S. M., Selvarajan, N., Hemalatha, S., & Rameshkumar, T. (2008). Hepatoprotective effect of Andrographis paniculata against cadmium-induced toxicity in male Wistar rats. *Toxicology International* 15(1), pp. (21-25).

Renugadevi, J., & Prabu, S. M. (2010). Cadmium-induced hepatotoxicity in rats and the protective effect of naringenin. *Experimental and Toxicologic Pathology*, 62(2), pp.171-181.

Renuka, M., Aparna, Y., Venkataramanaiah, P., & Srinivasulu Reddy, M. (2021). Vitamin C, E, and Zinc ameliorate Cadmium toxicity-induced biochemical changes in male albino rats. *Toxicology. Forensic Medical Journal*, *6*, pp. 13-19.

Sunitha, S., Nagaraj, M., & Varalakshmi, P. (2001). Hepatoprotective effect of lupeol and lupeol linoleate on tissue antioxidant defense system in cadmium-induced hepatotoxicity in rats. *Fitoterapia*, *72*(5), pp. 516-523.

Tietz, N. W. (1976). Fundamentals of clinical chemistry WB Saunders Co. *Philadelphia PA*, *47*.

Tosquy-Valle, O. H., Esqueda-Esquivel, V. A., Zetina-Lezama, R., & Ascencio-Luciano, G. (2010). Densidad y distancia de siembra en dos variedades de soya de temporal en Veracruz, México. *Agronomía Mesoamericana*, *21*(1), 63-72.

Unsal, V., Dalkıran, T., Çiçek, M., & Kölükçü, E. (2020). The role of natural antioxidants against reactive oxygen species produced by cadmium toxicity: a review. *Advanced Pharmaceutical Bulletin*, *10*(2), pp. 184.

Wendel, A. (1981). [44] Glutathione peroxidase. *In Methods in enzymology* (Vol. 77, pp. 325-333). Elsevier.

Williamson, E. M., Okpako, D. T., & Evans, F. J. (1996). Selection, *Preparation and Pharmacological Evaluation of Plant Material*, Volume 1 (Vol. 1). John Wiley & Sons.

Zaefarian, F., & Rezvani, M. (2016). Soybean (Glycine max [L.] Merr.) production under organic and traditional farming. *In Environmental stresses in Soybean production* pp. 103-129. Elsevier.