BF-1706
[1-8]

RESEARCH ARTICLE Indian Journal of Animal Research

Effect of a Combination of Citric Acid and Selenium Nanoparticles on Male Rats Nephrotoxicity Caused by Carbon Tetrachloride

Jameel Al-Tamimi[1], Hossam Ebaid[1], Iftekhar Hassan[1], Mohamed Habila[2], Nawal M. Al Malahi[1], Ibrahim M. Alhazza[1]

10.18805/IJAR.BF-1706

ABSTRACT

Background: To investigate the therapeutic effects of a combination of citric acid and Se-NPs against carbon tetrachloride (CCl_4)-induced renal damage in rats.

Methods: Twenty-four rats were divided into three groups (n = 8 each). Group 1 (control) received the vehicle, while Group 2 received a single dose of 1 ml/kg CCl_4 in liquid paraffin (1:1 volume) via intraperitoneal injection. Group 3 was administered a single dose of 1 ml/kg CCl_4 in liquid paraffin and then treated twice a week for three weeks with a dose of 2.5 mg/kg Se-NPs. One-way ANOVA and Tukey's method were used to compare the overall effects of each treatment.

Result: CCL_4 elevated MDA levels and decreased GSH levels in group 2 animals compared with those in the control group. Se-NPs significantly restored the oxidative stability in group 3. We found that CCL_4 significantly increased its effect on renal function, whereas Se-NPs restored the renal structure affected by CCL_4. As a result, Se-NPs can potentially reduce the markers of renal injury caused by CCl_4, while also restoring oxidative stability and renal structure and function.

Key words: CCl_4, Citric acid, Nephrotoxicity, Oxidative stress, Selenium nanoparticles.

INTRODUCTION

Globally, kidney disease or complications increase the risk of death to people with cardiovascular disease, diabetes, hypertension, age and obesity (Koye *et al.,* 2018). The prevalence of such renal disease ranges between 8% and 16% in patients with the mentioned diseases. Cellular damage may be caused by exposure to various organic molecules, including environmental contaminants and medications (Haghi *et al.,* 2014) that contribute to kidney diseases and nephrotoxicity (Bibu *et al.,* 2011; Sales and Foresto, 2020). Suppose the disease-induced, toxicant or drug-mediated nephrotoxicity is not addressed timely with adequate treatment. In that case, it can lead to severe health compromise, single or multiple organ failure, cancer and even death. Carbon tetrachloride (CCl_4) is an intensively researched chemical toxicant and environmental pollutant to study hepatotoxicity and nephrotoxicity *in vivo* (Baig and Khan, 2023). CCl_4 is commonly used as an organic solvent in dry cleaning, production of chloroform, aerosol propellant, fabric-spotting fluid, fire extinguisher fluid and refrigerating agent. However, during its usage in various forms, the compound has been established as a strong hepatoxicant and nephrotoxicant along with suspected carcinogen (Cohen *et al.,* 2023). Therefore, many investigators are working in this field of research to investigate in regulating its exposure to mankind and environment by administration of herbal active compound or drug (Ahmed *et al.,* 2020; Alhazza *et al.,* 2022; Alhazza *et al.,* 2020; Alhazza *et al.,* 2023; Ebaid *et al.,* 2021; Ebaid *et al.,* 2020; Hassan *et al.* 2019).

The use of nanotechnology for drug delivery may lead to the development of new types of medicines (Sahu *et al.,* 2021). Drug delivery is significantly affected by several biomolecules acting together. Extensive dataset analysis, interpretation methodologies and dataset applications for nanomaterials have improved and facilitated their use in health and environment sectors (Dixit *et al.,* 2022). Selenium nanoparticles (SeNPs) are extensively studied nanoparticles in dealing with various clinical and industrial issues (Ebaid *et al.,* 2021; Goltyaev and Varlamova, 2023). Selenium is essential trace element for the body because it

[1]Department of Zoology, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia.
[2]Department of Chemistry, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia.

Corresponding Author: Jameel Al-Tamimi, Department of Zoology, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia. Email: jhattamimi@gmail.com

How to cite this article: Al-Tamimi, J., Ebaid, H., Hassan, I., Habila, M., Al Malahi, N.M. and Alhazza, I.M. (2024). Effect of a Combination of Citric Acid and Selenium Nanoparticles on Male Rats Nephrotoxicity Caused by Carbon Tetrachloride. Indian Journal of Animal Research. DOI: 10.18805/IJAR.BF-1706.

Submitted: 22-09-2023 **Accepted:** 01-12-2023 **Online:** 04-01-2024

is a constituent of selenoproteins influencing the activity of glutathione peroxidase, immune cells and many other important metabolic enzymes and proteins. It also protects cells and tissues from free radicals mediated harm by functioning as an antioxidant (Geoffrion *et al.,* 2020). Selenium (Se) is often used as a dietary supplements (Nkengfack *et al.,* 2019; Skalickova *et al.,* 2017). However, depending on the chemical form of selenium, the period of supplementation and human health status, both a shortage and excess of selenium may be harmful to the body (Kieliszek and Bano, 2022). Selenium nanoparticles and citric acid have antioxidant, antibacterial and anticoagulant properties (Alhawiti, 2022).

Traditional selenium supplements have low absorption rates and high toxicity (Hosnedlova *et al.,* 2018). The bioavailability of this element increases at the nanoscale, allowing for its regulated release into the body (Hosnedlova *et al.,* 2018; Sadeghian *et al.,* 2012). Numerous studies have found that Se-NPs can be more effective in cancer chemoprevention as possible anticancer agents and drug delivery carriers (Zambonino *et al.,* 2023). Furthermore, its anti-metal toxicity properties are well established (Hassanin *et al.,* 2013). Additionally, the immunostimulatory action of nanoscale selenium has been found to positively affect various physiological activities (AbdEl-Kader *et al.,* 2023). Citric acid, in addition to its antioxidant bioactivity, promotes selenium stability (Abd El-Kader *et al.,* 2023).

As CCl_4- induced nephrpotoxicity is a serious medical concern, the present study is aimed to investigate whether supplementation with a combination of citric acid and selenium nanoparticles can effectively reduce CCl_4-mediated nephrotoxicity in rat animal model. Samples from the treated animals were subjected to basic biochemical analysis that were further confirmed by histological evaluation.

MATERIALS AND METHODS

Preparation of the combination of citric acid and selenium nanoparticles (Se-NPs)

As a selenium precursor, selenium dioxide was dissolved in water and thoroughly combined with citric acid by continuous stirring at room temperature for 3 h using a magnetic stirrer. Ascorbic acid (100 mL of 0.1 M solution) was then gently dropped. At 100°C, the reaction mixture was vigorously stirred for 10 hours. After centrifugation, selenium/citric acid nanocomposites were collected and analyzed by scanning electron microscopy (SEM) and energy-dispersive X-ray spectroscopy (EDS) (Khurana *et al.* 2019; Shahabi *et al.,* 2021).

Animals and experimental design

Twenty-four healthy male Swiss albino rats weighing 150-170 g (3-4 months old) were obtained from the Zoology Department (King Saud University, Riyad, Saudi Arabia). The rats acclimatized for a week to the laboratory settings in sufficiently large plastic cages before starting of the experiment. The rats were housed in an environment with a light-dark cycle of 12:12 h and a temperature range of 18-22°C, with access to food, water and ad libitum. The Animal Ethics Committee of King Saud University approved this study under ethical clearance number KSU-SE-20-38. Rats were divided into three groups (n = 8). Group 1 was treated with vehicle solution (Phosphate Buffered Saline) equivalent in volume to the other treatment groups and served as the control. Group 2 was administered a single intraperitoneal (IP) injection of 1 ml/kg of CCl_4 in liquid paraffin (1:1 v/v) (Makni *et al.,* 2011). After a single dose of CCl_4, group 3 was given at the dose of 2.5 mg/kg of the combination of citric acid and Se-NPs dissolved in phosphate buffer saline (PBS) as per a schedule of twice a week for three weeks (Ebaid *et al.,* 2021).

Blood and kidney samples

After anesthesia, blood was drawn by puncturing the heart directly and placing it in tubes without any anticoagulant. The blood was centrifuged at 3000 × g for 10 min and the serum was separated into Eppendorf tubes and stored at -20°C. Serum was used to determine levels of renal function as well as the amount of albumin. The kidneys were removed from sacrificed animals and washed with PBS. They were cut into two parts: one part of the kidney was used for histological study and the other was used to assess lipid peroxidation (MDA) and reduced glutathione (GSH).

Renal function and albumin tests

Renal function tests (urea and creatinine) and albumin levels were determined in serum samples. They were measured using commercial kits (Salucea Company, Netherlands) according to the manufacturer's instructions. A UV/visible spectrophotometer (Pharmacia Biotech, Cambridge, England) was used for all biochemical analyses.

Estimation of lipid peroxidation and an assay of reduced glutathione

Lipid peroxidation in the kidney was measured following the method described by (Ohkawa *et al.,* 1979) and the results were reported as nanomoles of MDA per mg of protein. Reduced glutathione was measured at 412 nm using the method described by (Moron *et al.,* 1979), with DTNB as the coloring reagent.

Histological study

Light microscopy was used to examine kidney samples to assess the effect of the treatment. The tissues were embedded in paraffin after fixation in 10% neutral-buffered formalin. Hematoxylin and eosin (H and E) staining was performed on the paraffin slices (H and E). Collagen deposits were stained with Mallory trichrome to separate the slides. Kidney damage was assessed by blind folding using a Leica DMRB/E light microscope (Switzerland). A rating score between (-: no change) and (+++: severe damage) was assigned to each investigated section (Dommels *et al.,* 2007).

Statistical analysis

Statistical analysis of the generated data was performed using SPSS software (IBM SPSS Statistics, Version 23). One-way analysis of variance (ANOVA) was used to determine the overall effects of each treatment. This analysis was supplemented by tests between treatments using Tukey's method for pairwise comparisons. Results are expressed as arithmetic mean (M)±standard deviation (SD), with $p<0.05$ as statistically significant between the treatment groups.

RESULTS AND DISCUSSION

Structural examination of Se-NPs/citric acid matrix

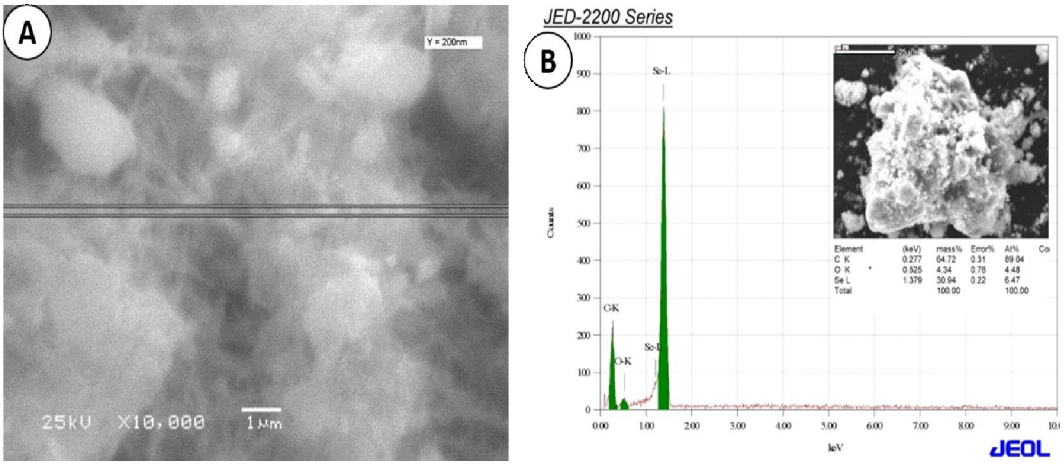
SEM examination of the prepared selenium/citric acid nanocomposite showed the heterogeneous shape of agglomerated needles and spheres with an average diameter of 200 nm (Fig 1). EDS examination showed the surface elements of carbon (64.72%), selenium (30.94%) and oxygen (4.34%), confirming the formation of citric acid in the nanocomposite. The combination of citric acid and selenium has many advantages for drug delivery. Selenium is an essential element that affects several physiological processes in the form of various selenoproteins. A moderate number of elements can prevent health problems by enhancing metabolism and boosting immunity (Hosnedlova *et al.,* 2018; Islam *et al.,* 2023; Sadeghian *et al.,* 2012). Hence, it contributes to a healthy life and increases average health. However, the element in its nanoform sheds its relative toxicity and is considered more appropriate for supplementation as a means of drug delivery or as an adjuvant to conventional medicine (Ferro *et al.,* 2021; Raza *et al.,* 2022).

Effect of the combination of citric acid and selenium nanoparticles on antioxidant status

Malondialdehyde (MDA) is a low-molecular-weight end product formed during the decomposition of some lipid peroxidation products. MDA estimation can be regarded as a diagnostic index for the extent of lipid peroxidation and oxidative injury in tissue lipids. Therefore, the present study considered MDA as a marker of lipid peroxidation induced by CCl_4. Group II showed a significant increase in MDA levels by (812.69%) compared to the control group I. However, in group III, the combination of citric acid and Se-NPs significantly restored the MDA level by (24.34%) concerning group II (Fig 2).

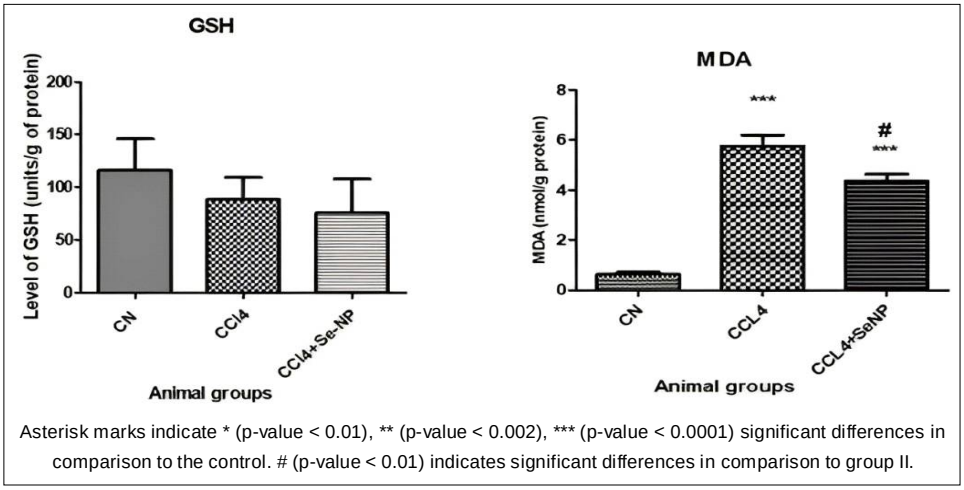
Reduced glutathione (GSH) acts as a scavenger and enzyme-catalyzed antioxidant in the oxidative injury of different tissues. Therefore, GSH plays a major role in protecting biological structures and functions. In the present study, reduced glutathione (GSH) was used as a primary parameter to determine the antioxidant status of the kidneys of treated animals. Group II showed a decrease in GSH level of (24.09%) in group I, while the CCl_4 + Se NP-treated group III showed a (13.90%) decrease in GSH level compared to group II (Fig 2).

Selenium is an essential component of many enzymes, some of which have antioxidant properties. Animals that are deficient in this element are more vulnerable to harm from certain forms of oxidative stress (Sobolev *et al.,* 2018). In this study, we investigated the effects of Se-NPs, a powerful antioxidant, on the rat's CCl_4-induced kidney damage and oxidative stress in an animal model. Our results indicated that Se-NPs exerted an ameliorative effect against CCl_4 nephrotoxicity. Our data confirm the formation of citric acid



(A) Scanning electron microscopy (SEM) of selenium nanoparticles showing the surface morphology at a magnification of 10000X. **(B)** Energy-dispersive X-ray spectroscopy (EDS) of Se nanoparticles.

Fig 1: The selenium nanoparticles were characterized by two prominent techniques.



Asterisk marks indicate * (p-value < 0.01), ** (p-value < 0.002), *** (p-value < 0.0001) significant differences in comparison to the control. # (p-value < 0.01) indicates significant differences in comparison to group II.

Fig 2: Bar diagram showing standard oxidative stress parameters [malondialdehyde (MDA) and reduced glutathione (GSH) in samples from different rat groups: group I (control) was treated with vehicle solution only, group II was treated with CCl_4 and group III was treated with CCl_4+Se NPs.

in the nanocomposite. The combination of citric acid and selenium has several advantages. These findings are consistent with those of a study conducted by Alhawiti, who found that citric acid and selenium nanoparticles have antioxidant, antibacterial and anticoagulant properties (Alhawiti, 2022). The combination of selenium and citric acid increases selenium stability and reduces the presence of the selenite form of the element, which is harmful and is associated with increased apoptosis (Fan *et al.,* 2020; Hosnedlova *et al.,* 2018; Nazıroğlu *et al.,* 2017; Qian *et al.,* 2019). Therefore, we planned this study to determine whether this combination could ameliorate CCl_4-induced renal toxicity *in vivo*. The combination of citric acid and Se-NPs replenished the activity of vital antioxidant enzymes and proteins, such as reduced glutathione in CCl_4 treated rats with greater efficacy.

Effect of the combination of citric acid and selenium nanoparticles on renal functions

Urea level is the most indicative marker of renal function. Interestingly, CCl_4 in Group II resulted in a significant increase in urea levels by (96.92%) compared to the control. However, the combination of citric acid and the combination of citric acid and Se-NPs in group III was found to demonstrate a decrease in its level by (20.31%) compared to group II (Fig 3). However, the urea concentration in Group III was still significantly higher than that in the control group.

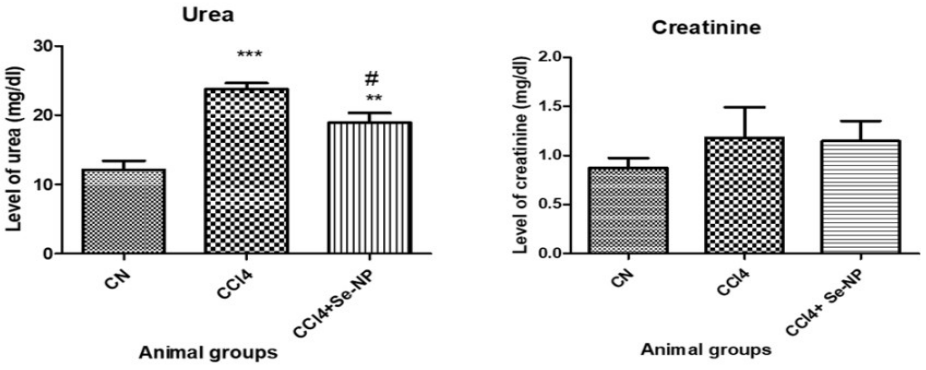
Creatinine level is an indicator of renal function. The creatinine level increased by (35.63%) in group II after CCl_4 injection. The combination of citric acid and Se-NPs decreased its level by (2.71%) in group III rats (Fig 3) compared to that in group II. However, no significant changes were observed in either group (II, III) compared with the control group. Statistical analysis revealed no significant changes in albumin concentrations during this study. However, its level was decreased by only (0.58%) in group II, while the combination of citric acid and Se-NPs enhanced it by (3.15%) in CCl_4 treated group III (Fig 4).

The critical parameters of kidney function (urea and creatinine), which were elevated in CCl_4 injected rats, tended towards normal values with the supplementation of the combination of citric acid and Se-NPs. Many previous studies have demonstrated a consistent relationship between nephrotoxicity and oxidative stress (Wu *et al.,* 2018). The results of our study on lipid peroxidation and renal function markers showed the same pattern (AlBasher *et al.,* 2020) reported that Se-NPs exhibit antioxidant properties. This study also confirmed that Se-NP supplementation could counter CCl_4-induced oxidative stress *in vivo*.

Kidney histology

Examination of the histopathological sections revealed that CCl_4 induced severe deterioration in renal tissues compared to control rats. Dilated urinary space with partially shrunken glomeruli was observed in the CCl_4 group. The combination of citric acid and Se-NPs showed a noticeable improvement in the renal tissues, exhibiting the urinary space and glomeruli structurally comparable to that of the control rats (Fig 5). Histological scores further confirmed that the combination of citric acid and Se-NPs noticeably improved the renal microstructure in the tissue samples in several histological indices; however, inflammatory cells were also observed in the same group (Table 1).

The kidney is one of the prime targets for xenobiotic and toxic substances because its large share of blood flow gives it the capacity to concentrate these substances (Timbrell and Barile, 2023). The glomerular hypertrophy in CCl_4-injected rats might be due to the proliferation of mesangial cells, which secrete more matrix as a defense mechanism. Furthermore, the blood capillaries appeared engorged with red blood cells and the urinary gap was obliterated in the kidney samples, as previously reported



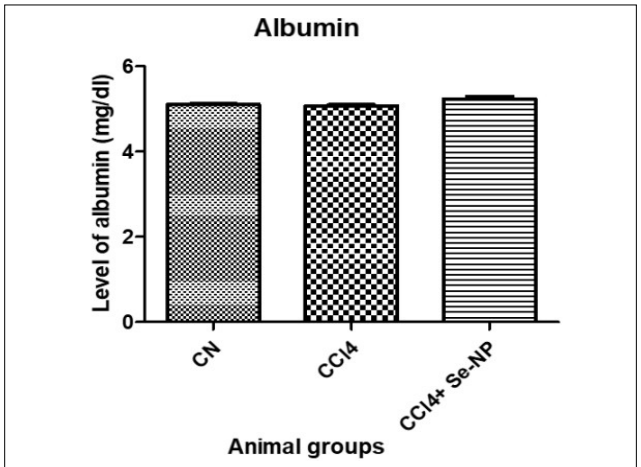
Asterisk marks indicate * (p-value < 0.01), ** (p-value < 0.002), *** (p-value < 0.0001) significant differences in comparison to the control. # (p-value < 0.01) indicates significant differences compared to group II.

Fig 3: Bar diagram showing renal function (urea and creatinine) in serum samples from different rat groups. Group I (control) was treated with vehicle solution only, group II was treated with CCl_4 and group III was treated with CCl_4+ Se NPs.

for indomethacin and piroxicam injections in rat animal models (Ebaid *et al.,* 2007; Good *et al.,* 2023). Infiltration of inflammatory cells into the intertubular tissues accompanied the tubular injury observed in the present study, possibly to minimize the lesion. Treatment with CCl_4 also induced the infiltration of inflammatory cells in the target organ and kidney, similar to the induction of inflammation by indomethacin and piroxicam in rat as animal models (Abd-Alla *et al.,* 2022).

Moreover, histological analysis of the tissue sections and their scores confirmed that considerable improvement was observed after the administration of NPs in the CCl_4 treated group. Here, the combination of citric acid and Se-NPs were found to lower lipid peroxidation and increase the level of GSH compared to control rats. Our study is in accord of previously reported by (Ansar *et al.,* 2017) showing supplementation with Se-NPs attenuates hepatotoxicity more strongly than supplementation with Ag-NPs. The combination of citric acid and Se-NPs increases plasma glutathione peroxidase (GPx) activity *in vivo* (Yanez-Lemus *et al.,* 2022). Therefore, our study is the first to report that combining SeNP with citric acid as nanocomposites are effective in ameliorating the CCl_4- induced nephrotoxicity.

In addition, they exhibited much lower toxicity when assessed based on the LD_{50}, acute kidney injury and short-term toxicity. The findings of the present study suggest that nanoscale selenium, such as Se-NPs with citric acid, can act as an antioxidant with a significantly reduced risk of toxicity compared with elemental selenium *in vivo* (Zambonino *et al.,* 2023). The effect of Se-NPs on selenoenzyme upregulation is equivalent to that of selenite and Se-NPs, with a considerable decrease in acute toxicity (Zambonino *et al.,* 2023).



Asterisk marks indicate * (p-value < 0.01), ** (p-value < 0.002), *** (p-value < 0.0001) significant differences in comparison to the control. # (p-value < 0.01) indicates significant differences compared to group II.

Fig 4: Bar diagram showing total serum albumin in different rat groups: group I (control) with vehicle solution only, group II treated with CCl_4 and group III treated with CCl_4+ Se-NPs.

Table 1: Histological score of the pathological changes in the renal tissues in rat treated only with CCL_4 and those treated with CCL_4 and Se-NPs.

Histopathological lesions	Oedematous glomeruli	Hemorrhage	Infiltration of inflammatory cells	Dilated glomerular space	Disintegrated nucleus
Control	-	-	-	-	-
CCL_4	+++	+++	+++	+++	+++
CCL_4 + Se-NPs	-	-	-	-	-

A rating score between (e: no change) and (+++: severe damage) was assigned for each investigated section (Dommels *et al.,* 2007).

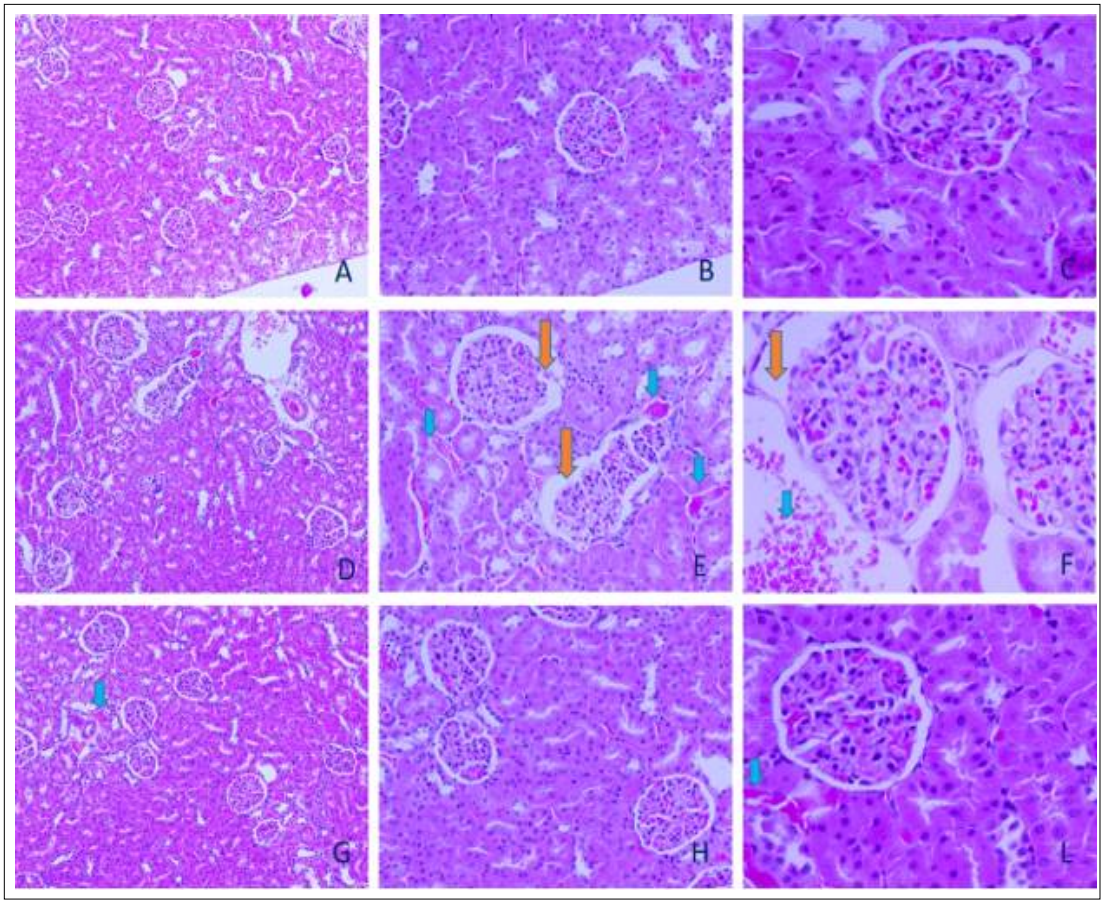


Fig 5: Representative microscopic images (H and E staining) at different magnifications for control, group I (A: 200X; B: 400X; C: 1000X); CCl_4 treated group II (D: 200X; E: 400X; F: 1000X) and CCl_4+Se-NP_S treated group III (G: 200X; H: 400X; L: 1000X). The renal tissues showed a dilated glomerular space in the CCl_4 rats (orange arrows) and dilated blood vessels with an observed hemorrhage.

CONCLUSION

In conclusion, this study confirmed that the combination of citric acid and Se-NPs alleviate CCl_4-induced renal toxicity and the histological structure of renal tissues. These findings will pave the way for new therapeutic horizons of Se as Se-NPs, either as a means of drug delivery or as an adjuvant to any established drug have nephrotoxicity as a major side effect. The study will benefit to the people who are exposed to CCl_4- based industries and consumer items. However, further studies are needed to understand the detailed mechanism involved in exploiting the advantages of Se-NPs with citric acid in biomedical and industrial applications.

ACKNOWLEDGEMENT

This work was supported by the Researchers Supporting Project number (RSP2024R366) of King Saud University, Riyadh, Saudi Arabia.

Conflict of interest

The authors declare that there are none.

REFERENCES

Abd-Alla, H.I., Ibrahim Fouad, G., Ahmed, K.A. and Shaker, K. (2022). Alloimperatorin from ammi majus fruits mitigates piroxicam-provoked gastric ulcer and hepatorenal toxicity in rats *via* suppressing oxidative stress and apoptosis. Biomarkers. 27(8): 727-742.

AbdEl-Kader, M.F., El-Kassas, S., Abd-Elghany, M.F., Abo-Al-Ela, H.G., El-Naggar, K., Al Wakeel, R.A., Zaki, A.G., Grana, Y.S. and El-Saftawy, H.A. (2023). Dietary selenium nanoparticles positively modulate the growth and immunity of seabream (*Sparus aurata*) fingerlings exposed to low salinity stress and vibrio parahaemolyticus challenge. Aquaculture. (576): 739893.

Ahmed, O.M., Ebaid, H., El-Nahass, E.S., Ragab, M. and Alhazza, I.M. (2020). Nephroprotective effect of pleurotus ostreatus and agaricus bisporus extracts and carvedilol on ethylene glycol-induced urolithiasis: Roles of Nf-Kb, P53, Bcl-2, Bax and Bak. Biomolecules 10(9): 1317.

AlBasher, G., Alfarraj, S., Alarifi, S., Alkhtani, S., Almeer, R., Alsultan, N., Alharthi, M., Alotibi, N., Al-Dbass, A. and Abdel Moneim, A.E. (2020). Nephroprotective role of selenium nanoparticles against glycerol-induced acute kidney injury in rats. Biological Trace Element Research. 194(2): 444-454.

Alhawiti, A.S. (2022). Citric acid-mediated green synthesis of selenium nanoparticles: Antioxidant, antimicrobial and anticoagulant potential applications. Biomass Conversion and Biorefinery. pp. 1-10.

Alhazza, I.M., Ebaid, H., Omar, M.S., Hassan, I., Habila, M.A., Al-Tamimi, J. and Sheikh, M. (2022). Supplementation with selenium nanoparticles alleviates diabetic nephropathy during pregnancy in the diabetic female rats. Environmental Science and Pollution Research. 29(4): 5517-5525.

Alhazza, I.M., Hassan, I., Ebaid, H., Al-Tamimi, J. and Alwasel, S.H. (2020). Chemopreventive effect of riboflavin on the potassium bromate-induced renal toxicity *in vivo*. Naunyn-Schmiedeberg's Archives of Pharmacology. 393(12): 2355-2364.

Alhazza, I.M., Hassan, I., Ebaid, H., Al-Tamimi, J. and Hasan, Z. (2023). Zinc oxide nanoparticles blunt potassium-bromate-induced renal toxicity by reinforcing the redox system. Molecules. 28(13): 5084.

Ansar, S., Alshehri, S.M., Abudawood, M., Hamed, S.S. and Ahamad, T. (2017). Antioxidant and Hepatoprotective Role of Selenium against Silver Nanoparticles. International Journal of Nanomedicine 12: 7789.

Baig, M.F. and Khan, M.K.A. (2023). Renal toxicity induced by carbon tetrachloride in experimental model: Renal toxicity induced by ccl4 in experimental model. Pakistan BioMedical Journal. pp. 30-35.

Bibu, K., Joy, A., Abraham, M.J. and Mercey, K. (2011). Evaluation of therapeutic effect of aqueous extract of stem bark of mangifera indica linn. On gentamicin-induced nephrotoxicity. Journal of Applied Animal Research. 39(3): 252-256.

Cohen, S.M., Bevan, C., Gollapudi, B. and Klaunig, J.E. (2023). Evaluation of the carcinogenicity of carbon tetrachloride. Journal of Toxicology and Environmental Health. Part (B), pp. 1-29.

Dixit, R., Khambhati, K., Supraja, K.V., Singh, V., Lederer, F., Show, P.L., Awasthi, M.K., Sharma, A. and Jain, R. (2022). Application of machine learning on understanding biomolecule interactions in cellular machinery. Bioresource Technology. p. 128522.

Dommels, Y., Butts, C., Zhu, S., Davy, M., Martell, S., Hedderley, D., Barnett, M., McNabb, W. and Roy, N. 2007. Characterization of intestinal inflammation and identification of related gene expression changes in Mdr1a"/" Mice. Genes and Nutrition 2(2): 209-223.

Ebaid, H., Al-Tamimi, J., Hassan, I., Habila, M.A., Rady, A.M., Alhazza, I.M. and Ahmed, A M. (2021). Effect of selenium nanoparticles on carbon tetrachloride-induced hepatotoxicity in the swiss albino rats. Applied Sciences. 11(7): 3044.

Ebaid, H., Bashandy, S.A., Abdel-Mageed, A.M., Al-Tamimi, J., Hassan, I. and Alhazza, I.M. (2020). Folic acid and melatonin mitigate diabetic nephropathy in rats via inhibition of oxidative stress. Nutrition and Metabolism. 17(1): 1-14.

Ebaid, H., Dkhil, M.A., Danfour, M.A., Tohamy, A. and Gabry, M.S. (2007). Piroxicam-induced hepatic and renal histopathological changes in mice. Libyan Journal of Medicine. 2(2): 82-89.

Fan, D., Li, L., Li, Z., Zhang, Y., Ma, X., Wu, L., Zhang, H. and Guo, F. (2020). Biosynthesis of selenium nanoparticles and their protective, antioxidative effects in streptozotocin induced diabetic rats. Science and Technology of Advanced Materials. 21(1): 505-514.

Ferro, M., Charneca, S., Dourado, E., Guerreiro, C.S. and Fonseca, J.E. (2021). Probiotic supplementation for rheumatoid arthritis: A promising adjuvant therapy in the gut microbiome era. Frontiers in Pharmacology. 12: 711788

Geoffrion, L.D., Hesabizadeh, T., Medina-Cruz, D., Kusper, M., Taylor, P., Vernet-Crua, A., Chen, J., Ajo, A., Webster, T.J. and Guisbiers, G. (2020). Naked selenium nanoparticles for antibacterial and anticancer treatments. ACS Omega. 5(6): 2660-2669.

Goltyaev, M.V. and Varlamova, E.G. (2023). The role of selenium nanoparticles in the treatment of liver pathologies of various natures. International Journal of Molecular Sciences. 24(13): 10547.

Good, P.I., Li, L., Hurst, H.A., Herrera, I.S., Xu, K., Rao, M., Bateman, D.A., Al-Awqati, Q., D'Agati, V.D. and Costantini, F. (2023). Low nephron endowment increases susceptibility to renal stress and chronic kidney disease. JCI Insight. 8(3): e161316.

Haghi, M.E., Dehghan, G., Banihabib, N., Zare, S., Mikaili, P. and Panahi, F. (2014). Protective effects of cornus mas fruit extract on carbon tetrachloride induced nephrotoxicity in rats. Indian Journal of Nephrology. 24(5): 291.

Hassan, I., Husain, F.M., Khan, R.A., Ebaid, H., Al-Tamimi, J., Alhazza, I.M., Aman, S. and Ibrahim, K.E. (2019). Ameliorative effect of zinc oxide nanoparticles against potassium bromate-mediated toxicity in swiss albino rats. Environmental Science and Pollution Research. 26(10): 9966-9980.

Hassanin, K.M., Abd El-Kawi, S.H. and Hashem, K.S. (2013). The prospective protective effect of selenium nanoparticles against chromium-induced oxidative and cellular damage in rat thyroid. International Journal of Nanomedicine. 8: 1713.

Hosnedlova, B., Kepinska, M., Skalickova, S., Fernandez, C., Ruttkay-Nedecky, B., Peng, Q., Baron, M., Melcova, M., Opatrilova, R. and Zidkova, J. (2018). Nano-selenium and its nanomedicine applications: A critical review. International Journal of Nanomedicine. 13: 2107.

Islam, M.R., Akash, S., Jony, M.H., Alam, M.N., Nowrin, F.T., Rahman, M.M., Rauf, A. and Thiruvengadam, M. (2023). Exploring the potential function of trace elements in human health: A therapeutic perspective. Molecular and Cellular Biochemistry. pp. 1-31.

Khurana, A., Tekula, S., Saifi, M.A., Venkatesh, P. and Godugu, C. (2019). Therapeutic applications of selenium nanoparticles. Biomedicine and Pharmacotherapy. 111: 802-812.

Kieliszek, M. and Bano, I. (2022). Selenium as an important factor in various disease states: A review. EXCLI Journal. 21: 948-966.

Koye, D.N., Magliano, D.J., Nelson, R.G. and Pavkov, M.E. (2018). The global epidemiology of diabetes and kidney disease. Advances in Chronic Kidney Disease. 25(2): 121-132.

Makni, M., Chtourou, Y., Fetoui, H., Garoui, E.M., Boudawara, T. and Zeghal, N. (2011). Evaluation of the antioxidant, anti-inflammatory and hepatoprotective properties of vanillin in carbon tetrachloride-treated rats. European Journal of Pharmacology. 668(1-2): 133-139.

Moron, M.S., Depierre, J.W. and 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): 67-78.

Nazýroðlu, M., Muhamad, S. and Pecze, L. (2017). Nanoparticles as potential clinical therapeutic agents in alzheimer's disease: Focus on selenium nanoparticles. Expert Review of Clinical Pharmacology. 10(7): 773-782.

Nkengfack, G., Englert, H. and Haddadi, M. (2019). Selenium and immunity. Nutrition and Immunity. pp. 159-165.

Ohkawa, H., Ohishi, N. and Yagi, K. (1979). Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Analytical Biochemistry. 95(2): 351-358.

Qian, F., Misra, S. and Prabhu, K.S. (2019). Selenium and selenoproteins in prostanoid metabolism and immunity. Critical Reviews in Biochemistry and Molecular Biology. 54(6): 484-516.

Raza, A., Johnson, H., Singh, A. and Sharma, A.K. (2022). Impact of selenium nanoparticles in the regulation of inflammation. Archives of Biochemistry and Biophysics. p. 109466.

Sadeghian, S., Kojouri, G.A. and Mohebbi, A. (2012). Nanoparticles of selenium as species with stronger physiological effects in sheep in comparison with sodium selenite. Biological Trace Element Research. 146(3): 302-308.

Sahu, T., Ratre, Y.K., Chauhan, S., Bhaskar, L., Nair, M.P. and Verma, H.K. (2021). Nanotechnology based drug delivery system: Current strategies and emerging therapeutic potential for medical science. Journal of Drug Delivery Science and Technology. 63: 102487.

Sales, G.T.M. and Foresto, R.D. (2020). Drug-induced nephrotoxicity. Revista da Associação Médica Brasileira. 66: 82-90.

Shahabi, R., Anissian, A., Javadmoosavi, S.A. and Nasirinezhad, F. (2021). Protective and anti-inflammatory effect of selenium nano-particles against bleomycin-induced pulmonary injury in male rats. Drug and Chemical Toxicology. 44(1): 92-100.

Skalickova, S., Milosavljevic, V., Cihalova, K., Horky, P., Richtera, L. and Adam, V. (2017). Selenium Nanoparticles as a nutritional supplement. Nutrition. 33: 83-90.

Sobolev, O., Gutyj, B., Petryshak, R., Pivtorak, J., Kovalskyi, Y., Naumyuk, A., Petryshak, O., Semchuk, I., Mateusz, V. and Shcherbatyy, A. (2018). biological role of selenium in the organism of animals and humans. Ukrainian Journal of Ecology. 8(1): 654-665.

Timbrell, J. and Barile, F.A. (2023). Introduction to Toxicology. CRC Press. https://doi.org/10.1201/9781003188575.

Wu, Q., Wang, X., Nepovimova, E., Wang, Y., Yang, H. and Kuca, K. (2018). Mechanism of cyclosporine a nephrotoxicity: Oxidative stress, autophagy and signalings. Food and Chemical Toxicology. 118: 889-907.

Yanez-Lemus, F., Moraga, R., Mercado, L., Jara-Gutierrez, C., Smith, C.T., Aguayo, P., Sanchez-Alonzo, K., García-Cancino, A., Valenzuela, A. and Campos, V.L. (2022). Selenium nanoparticles biosynthesized by pantoea agglomerans and their effects on cellular and physiological parameters in the rainbow trout oncorhynchus mykiss. Biology. 11(3): 463.

Zambonino, M.C., Quizhpe, E.M., Mouheb, L., Rahman, A., Agathos, S.N. and Dahoumane, S.A. (2023). Biogenic Selenium nanoparticles in biomedical sciences: Properties, current trends, novel opportunities and emerging challenges in theranostic nanomedicine. Nanomaterials. 13(3): 424.