Official Journal Issued by Faculty of Veterinary Medicine

Benha Veterinary Medical Journal

Journal homepage: https://bvmj.journals.ekb.eg/

Since 1990

Original Paper

Ameliorative activity of cinnamon extract (*Cinnamomum Cassia*) against aluminum oxide nanoparticle (Al_2O_3 NP) induced hepatic damage

Ebtssam Beder [1]*, Hatem Bakry [1], Mohamed Abosalem [1], Ahmed Abdeen[1], Sara Badawy[2]

[1] *Department of Forensic Medicine and Toxicology, Faculty of Veterinary Medicine, Benha University, Toukh 13736, Egypt.*

[2] *Department of Pathology, Faculty of Veterinary Medicine, Benha University, Toukh 13736, Egypt*

ARTICLE INFO

Keywords

Al_2O_3NP

Antioxidants

Cinnamon

Hepatotoxicity

Residues

Received 09/10/2024
Accepted 01/11/2024
Available On-Line
xx/xx/2024

ABSTRACT

Aluminum oxide nanoparticle (Al_2O_3NP) is a common Life-threatening environmental pollutant. It performs its toxic action via induction of lipid peroxidation, protein oxidation, disruption of the Redox system, and finally cellular death. Therefore, this study aimed to find out whether cinnamon may protect rats from liver damage caused by Al_2O_3NP. Forty male Wister albino rats were categorized into four Similar groups at random. The control group was given Milli-Q water orally for 28 days, cinnamon group was given 200 mg/kg/day of cinnamon disintegrated in Milli-Q water orally for 28 days, Al_2O_3NP group was given 100 mg/kg/day of Al_2O_3NP disintegrated in Milli-Q water orally for 28 days. Al_2O_3NP + cinnamon group was given 100 mg/kg/day of Al_2O_3NP, and 200 mg/kg/day of cinnamon dissolved in Milli-Q water orally for 28 days. Al_2O_3NP exposed rats' serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and total cholesterol (TC) dramatically increased. Moreover, decreased glutathione reduced (GSH), while malondialdehyde (MDA) and hydrogen peroxide (H_2O_2) levels in the hepatic tissues significantly increased. Histopathological examination of liver sections confirmed this hepatic damage where hepatic degeneration and necrosis were observed. Cinnamon co-administration was effective in restoring the parameters to normal and reducing hepatic injury in Al_2O_3NP-intoxicated rats. As a result, our findings indicate that cinnamon may have protective effects due to its antioxidant defense systems.

1. INTRODUCTION

Nanoparticles find applications across various sectors, including industry, defense, and healthcare. In the field of nanotechnology, the tools or materials created typically measure between 1 and 100 nanometers (Sajid et al., 2015). Besides their diminutive size, NPs are readily adopted by cells and can penetrate the bloodstream and lymphatic systems. They could settle in different tissues and organs, but this buildup can cause toxic effects (M'rad et al., 2018). Aluminum oxide nanoparticle (Al_2O_3NP) is commonly utilized in cosmetics and other skin care manufactured products (Poborilova et al., 2013). Interestingly, the exposure Al_2O_3NP may cause the cells to produce reactive oxygen species (ROS) and diminish antioxidant activity levels (Shrivastava et al., 2014). Moreover, Al_2O_3NP causes bioaccumulation in certain organs like kidneys, liver, testis, and brain which has negative environmental implications (Stanley et al., 2010).

The cinnamon plant (*Cinnamomum Cassia*) with the generic name Cinnamon is a widely used medicinal herb in traditional medicine and is also used to flavor meals and pastries (Sakr and Albarakai, 2014). Cinnamon is demonstrated to act as an antioxidant, anti-inflammatory, anticancer, antibacterial, antidiabetic, lipid-lowering, and cardiovascular-disease-lowering substance (Rao and Gan, 2014). The antioxidant activity of cinnamon could result from scavenging ROS, boosting antioxidant defense system activity, and reducing peroxidation of lipids (Abdeen et al., 2017).

This study's objective was to investigate the possible potential benefit of cinnamon on Al_2O_3NP-induced hepatotoxicity in rats by evaluating its role in controlling oxidative injury.

2. MATERIAL AND METHODS

2.1. Chemicals:

Aluminum oxide nanoparticle (Al_2O_3NP) of less than 50 nm in particle size was acquired from Sigma-Aldrich (Germany). Cinnamon barks were purchased from AG International for Import and Export (Nasr City, Cairo, Egypt). The analytical kits were bought from SPINREACT (Girona, Spain).

2.2. Plant extract preparation:

The dried cinnamon barks were used after grinding by the homogenizer to prepare an alcoholic plant-based extract. Briefly, 100 g of the desiccated grinded bark was soaked in one Liter of alcohol 70 % (distilled water: absolute ethanol, 70:30, v/v) for 72 hours followed by filtration. The filtrate was then heated for an entire night at 70 °C using a water bath. After being dried, the extract was weighed and stored for additional examinations and use (Harborne, 1973; Zhang et al., 2007).

* Correspondence to: ebtssam.beder@fvtm.bu.edu.eg

2.3. Experimental design:
Forty male Wister albino rats with an average weight of 160 ± 10 g were obtained from the Institute of Veterinary Serum and Vaccine Research, Abasia, Cairo, Egypt. The experimental rats followed the instructions for the use and management of laboratory animals that have been granted ethical approval by the Research Ethical Committee of Benha University, Faculty of Veterinary Medicine, Egypt (Approval No. BUFVTM 04-09-23). Rats were weighed weekly and split at random into four groups as follows:

- Control group: 10 rats were given Milli-Q water orally for 28 days.
- Cinnamon group: 10 rats were given 200 mg/kg b. wt. per day of cinnamon dissolved in Milli-Q water orally for 28 days (Sakr and Albarakai, 2014; Abdeen et al., 2017).
- Al_2O_3NP group: 10 rats were given 100 mg/kg b. wt. per day of Al_2O_3NP dissolved in Milli-Q water orally for 28 days (Abou-Zeid et al., 2021; El-Borai et al., 2022).
- Al_2O_3NP + cinnamon group: 10 rats were given 100 mg/kg b. wt. per day of Al_2O_3NP and 200 mg/kg b. wt. per day of cinnamon dissolved in Milli-Q water orally for 28 days.

Aluminum oxide nanoparticle was freshly dispersed daily in Milli-Q water by vigorous vortexing for 30 min using an electrical shaker to provide an even and consistent dispersion of particles. One day following the last dose, isoflurane 100% was used to anesthetize rats.

2.4. Sampling:
Blood samples were collected from Vena Cava at the end of the experiment. Then all rats were euthanized, and liver tissues were taken out and cut into two parts, one for histopathological investigation, and the other was preserved at -80 °C for oxidative stress markers analysis.

2.5. Serum biochemical analysis:
After centrifugation at 3000 rpm for 15 minutes, clear sera were recovered, then refrigerated at -20 °C for subsequent biochemical examination, which included aspartate aminotransferase (AST), alanine aminotransferase (ALT) (Huang et al., 2006), Alkaline phosphatase (ALP) (Rosalki and Foo, 1984), and Total cholesterol (TC) (Allain et al., 1974). All values were measured by using a spectrophotometer, JASCO 7800, un/vis, JAPAN according to the data protocol provided by using analytical kits obtained from SPINREACT (Girona, Spain).

2.6. Evaluation of oxidative stress markers:
The liver samples were homogenized by using an electrical homogenizer (COLOMBIA INTERNATIONAL) at speed 6 for one minute where 300 mg tissue was homogenized with 3 ml phosphate buffer saline pH 7.4, After proper homogenization, the homogenates were centrifuged at 4000 rpm for 15 minutes at 4 °C by using a cooling centrifuge for separation of supernatants. These supernatants were used for the detection of oxidative stress biomarkers. Oxidative stress biomarkers were glutathione reduced (GSH) (Sezgintürk and Dinçkaya, 2011), malondialdehyde (MDA) (Giera et al., 2012), and hydrogen peroxide (H_2O_2) (Werner, 2003) using commerical kits (Biodiagnostic Co., Egypt).

2.7. Histopathological examination:
The liver tissue was preserved in 10% neutral buffered formalin for 72 hours. After proper fixation, the liver tissue was fixed in paraffin, cut into 5 µm thickness slices, and stained with hematoxylin and eosin (H&E) for histopathological examination according to the method described by Bancroft and Gamble (2008). Histopathological changes were examined with a Nikon Eclipse E800 light microscope (Melville, NY, USA) and images were captured with an Olympus digital camera.

2.8. Microwave digestion of liver tissue and assessment of aluminum residue by ICP-MS:
4.5 mL of 69% HNO_3, 0.5 mL H_2O_2, and 250 mg of liver sample were digested in a Milestone microwave digestion system (ETHOS UP, Italy) by heating to 200 °C, over 20 min and maintaining this temperature for 20 min. Then cooling occurs to prevent loss of volatile analytics. Post digestion, about 2 ml of extract was diluted by 8 ml milli-Q water. This solution was subjected to ICP-MS/MS (iCAP-ICP-MS, Thermofilm, Germany) analysis as described by Korotkova et al. (2022).

2.9. Statistical analysis:
Using SPSS software (Version 26; IBM, Chicago, USA), the collected data was statistically evaluated using a one-way ANOVA and Duncan's multiple range test for post hoc analysis to find out the significance between the various groups at $P \leq 0.05$. Values were presented as mean ± SE.

4. RESULTS

3.1. Biochemical analysis:
Administration of Al_2O_3NP significantly elevated the levels of AST, ALP, and total cholesterol as well as ALT. The co-administration of cinnamon extract showed a significant decrease in these parameters compared to Al_2O_3NP group as shown in Fig. (1).

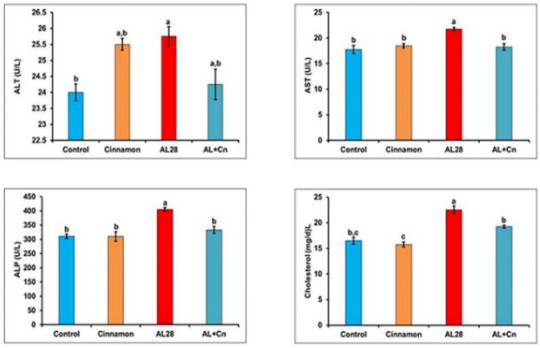

Figure 1 Serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and total cholesterol (TC) in control and treated rats. The data are expressed as Mean ± SE. There is a significant difference at ($P \leq 0.05$) between mean values in the same row that have different superscript letters.

3.2. Oxidative stress markers assessment:
Liver damage caused by Al_2O_3NP led to a dramatic decrease in GSH level with remarkable elevation in MDA and H_2O_2 activities in liver tissue compared to the control group. When Al_2O_3NP intoxicated rats were treated with cinnamon, GSH was significantly increased while MDA and H_2O_2 were restored to normal levels in the hepatic tissue when compared to the Al_2O_3NP group as shown in Fig. (2).

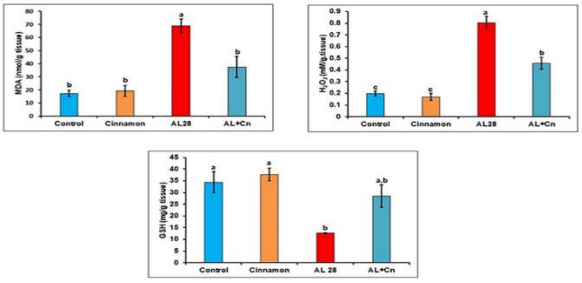

Figure 2 Liver oxidative stress biomarkers levels of glutathione reduced (GSH), malondialdehyde (MDA) and hydrogen peroxide (H_2O_2) in control and treated rats. The data are expressed as Mean ± SE. There is a significant difference at ($P \leq 0.05$) between mean values in the same row that have different superscript letters.

3.3. Histopathological findings:

The microscopical examination of liver sections from the control and Cn groups displayed normal histological appearance of the central vein, blood sinusoids, hepatic cells as well as the portal area (Fig. 3A&3B). In contrast, rats intoxicated with Al_2O_3NP for 28 days exhibited notable vascular alterations characterized by congested and dilated central veins and hepatic sinusoids (Fig. 3C) with activated Kupffer cells (Fig. 3D). A recent thrombus formation was observed in the central vein (Fig. 3C). Most hepatocytes showed marked degeneration and coagulative necrosis with pyknotic nuclei (Fig. 3D). Besides, focal aggregation of mono-nuclear leucocytic cellular infiltration was detected in the hepatic parenchyma (Fig. 3E). The majority of periportal hepatocytes revealed fatty changes (Fig. 3G). Interestingly, there was a mild degree of periductal fibrosis with multiple newly formed bile ductules (Fig. 3F). Meanwhile, the majority of the liver sections treated with Cn showed almost normal hepatic architecture with only mild degeneration and a marked reduction in hepatic inflammation and fibrosis induced by Al_2O_3NP (Fig. 3H).

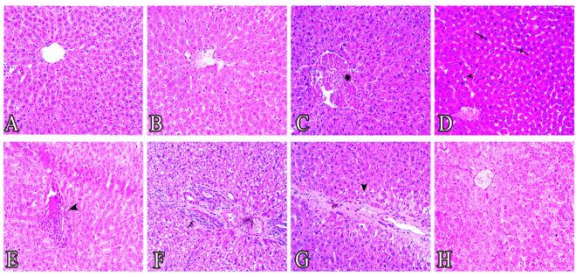

Figure 3 Representative photomicrographs of H&E stained-liver tissue. (A) control, (B) Cn group revealing normal hepatic histological architecture. (C-G) Al-intoxicated rats showing (C) marked dilation, congestion and thrombus (asterisk) formation in central vein, (D) extensive hepatic necrosis with pyknotic nuclei (arrow) and dilated hepatic sinusoids with activated Kupffer cells (arrowhead), (E) focal mononuclear leucocytic cellular infiltration mainly lymphocytes (arrowhead), (F) portal and periportal inflammation and fibrosis (arrow) with newly formed bile ductulus (G) periportal fibrous connective tissue proliferation and periportal fatty changes (arrowhead). (H) Cn+ Al group revealing near normal histological structure of liver with mild hepatic degeneration×200.

3.8. Effect of cinnamon on aluminum residue in liver tissues:

According to Fig. (4), Samples and standards were contrasted within the linear range of the calibration. The correlation coefficient of Al was found to be 0.9294. There was a significant increase in Al residues in Al_2O_3NP group (27000 ppb) relative to the control group, while cinnamon supplementation significantly decreased Al accumulation in liver tissue in comparison to the Al_2O_3NP group ($P \leq 0.05$).

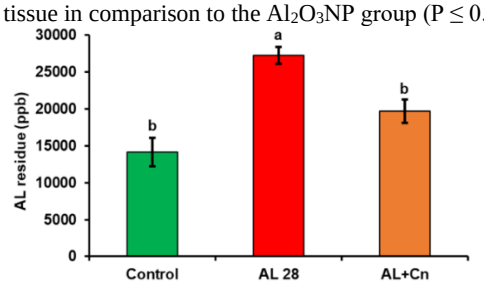

Figure 4. Al residues showed a significant increase in Al_2O_3NP group in relative to control group, while cinnamon supplementation significantly decreases Al accumulation in liver tissue in comparison to the Al_2O_3NP group ($P \leq 0.05$).

4. DISCUSSION

Nanotechnology is a recent scientific field focusing on producing items with unique qualities by creating and modifying nanoparticles (NPs), which have a size range of 1 to 100 nm (Yousef et al., 2019). As a result, there is a higher chance that NPs will interact with cells, and their effects on the body differ from those of common particle pollutants. Depending on the materials utilized, NPs' toxicity may increase significantly when inducing cytotoxicity due to their huge surface area (Li et al., 2020).

The results of the current investigation demonstrated that oral administration of Al_2O_3NP caused a considerable alteration in several liver biomarkers and histological parameters. Al_2O_3NP can easily pass biological barriers and accumulate in many organs and tissues due to their tiny size and strong surface reactivity (Krause et al., 2020). Following their exposure, these particles are attracted to the inside of cells through electrostatic interactions as Al_2O_3NP is positively charged and the cell surface is negatively charged (Liu et al., 2020).

According to Silva et al. (2005), Al_2O_3NP accumulation inside the hepatocytes may change the metabolic pathway of phosphate and ATP. This may result in cellular energy depletion, a disruption of the membrane potential that could lead to necrosis (as shown by our histological analysis), leading to a loss of cell membrane integrity, increased permeability, and release of transaminases into the bloodstream (Nehru and Anand, 2005). The direct correlation between the accumulation of Al_2O_3NP and liver MDA levels supports the idea that Al_2O_3NP enhanced lipid oxidation by ROS which increases the liberation of AST and ALT into ~~the~~ circulation (Morsy, El-Ala, et al., 2016). These findings corroborate those of El-Borai et al. (2022) and Makwana et al. (2024), who found that rats treated with Al_2O_3NP had significantly higher serum levels of ALT and AST. According to Makwana et al. (2024) and Yousef et al. (2019) Al_2O_3NP led to a dramatic increase in ALP which is considered one of the hepatic-induced enzymes (ALP) which is consistent with the current findings. Rats intoxicated with Al_2O_3NP had higher levels of total cholesterol (TC) as a result of lipid peroxidation which matched with those of Yousef et al. (2022).

Previous studies demonstrated that hepatic GSH, MDA, and H_2O_2 are potential targets for Al_2O_3NP toxicity. Our findings showed a dramatic reduction in the level of hepatic GSH in the toxic group compared to the control group which matched with those of El-Borai et al. (2022) and Yousef et al. (2022). Moreover, in the current investigation, we found that the rats treated with Al_2O_3NP displayed a significant increase in the level of MDA as a result of severe lipid peroxidation of the hepatocytes caused by overproduction of ROS which corroborated those of Morsy, Abou el-Ala et al. (2016); Yousef et al. (2019). According to Kiruthiga et al. (2007), H_2O_2 is the major reactive oxygen species generated During the hepatic detoxification of almost all environmental contaminants, H_2O_2 dramatically increased in the intoxicated rats compared to the control animals as a result of hepatocellular injury and oxidative damage.

In our investigation, cinnamon administration given orally considerably reduced alterations in the biochemical and oxidative parameters and kept the values of the parameters under evaluation close to normal. Increased activity of ALT, AST, ALP In the serum of rats treated with Al_2O_3NP was normalized in the serum of cinnamon treated Al_2O_3NP group. The strong antioxidant properties of cinnamon may be the cause of the drop in serum enzyme activity (Hussain et al., 2020). In hypercholesterolemic albino rats, cinnamon extract has hypolipidemic effects. When the extract was given daily, total cholesterol (TC) levels in the blood were lower than in the intoxicated group (Błaszczyk et al., 2021). Our study revealed that cinnamon showed a notable increase in the activity of antioxidant enzymes like GSH. Additionally, Cinnamon extract co-administration had a protective effect through decreasing MDA levels compared to the toxic group. Because of its high phenolic and

flavonoid content, cinnamon may have a suppressive impact on the production of ROS, protecting against Al_2O_3NP-induced liver damage (Azab et al., 2011). Moreover, cinnamon extract contains significant levels of phenolic compounds and shows possible hydrogen peroxide (H_2O_2) blocking ability compared to intoxicated rats (Rao and Gan, 2014).

The liver damage caused by Al_2O_3NP was verified in further detail by histological analysis. In the current research, histopathological findings revealed that the oral administration of Al_2O_3NP induced vascular alterations such as congestion and dilation of central veins and hepatic sinusoids which may be attributed to Al_2O_3NP ability to trigger the expression of inflammatory molecules, such as intercellular adhesion molecule-1, interleukin-8, monocyte chemotactic protein-1 and various adhesion molecules, resulting in endothelial damage (Gojova et al., 2007). The current findings were consistent with those of Abo-EL-Sooud et al. (2023) and Makwana et al. (2024), who reported that Al_2O_3NP causes congestion of blood vessels and sinusoids, vasculitis, and hypertrophy of blood vessel wall. Endothelial dysfunction is the primary predisposing factor in thrombus formation, as revealed in the current investigation. Moreover, hepatic degeneration and necrosis may result from the overproduction of ROS induced by Al_2O_3NP (Siddique et al., 2011). Similar pathological alterations in the liver were reported by Abo-EL-Sooud et al. (2023); Alghriany et al. (2022) and Makwana et al. (2024). The recorded mild portal fibrosis was a sequel to the inflammation induced by Al_2O_3NP (Abo-EL-Sooud et al. 2023). However, in our study, Cn co-treatment improved the pathological alterations induced by Al_2O_3NP and significantly attenuated hepatic inflammation and fibrosis due to its phenolic and flavonoid levels, which have a suppressive impact on the production of ROS (Azab et al. 2011) and its anti-inflammatory effect (Hussain et al. 2020; Hussain et al., 2021). Also, the current findings were agreed with Elkomy et al. (2016) and Hussain et al. (2020). Additionally, Al residues showed a dramatic increase in Al_2O_3NP group in relative to the control group, while cinnamon supplementation significantly decreases Al accumulation in liver tissue in comparison to the Al_2O_3NP group.

5. CONCLUSIONS

The results of the current investigation showed that cinnamon antioxidant and anti-inflammatory properties alleviate oxidative damage of Al_2O_3NP-induced hepatic injury via amelioration of all hepatic biomarkers and pathological changes.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this article.

ACKNOWLEDGMENTS

All authors appreciate the technical support provided by the staff members of the Department of Forensic Medicine and Toxicology, Faculty of Veterinary Medicine, Benha University, for implementing this study.

6. REFERENCES

1. Abdeen, A., Ghonim, A., El-Shawarby, R., Abdel-Aleem, N., El-Shewy, E., Abdo, M., and Abdelhiee, E. 2017. Protective effect of cinnamon against cadmium-induced hepatorenal oxidative damage in rats. International Journal of Pharmacology and Toxicology, 5,1, 17–22. https://doi.org/10.14419/ijpt.v5i1.7119
2. Abo-EL-Sooud, K., Abd-Elhakim, Y. M., Hashem, M. M. M., El-Metwally, A. E., Hassan, B. A., and El-Nour, H. H. M. 2023. Ameliorative effects of quercetin against hepatic toxicity of oral sub-chronic co-exposure to aluminum oxide nanoparticles and lead-acetate in male rats. Naunyn-Schmiedeberg's Archives of Pharmacology, 396,4, 737–747. https://doi.org/10.1007/s00210-022-02351-y
3. Abou-Zeid, S. M., Elkhadrawey, B. A., Anis, A., AbuBakr, H. O., El-Bialy, B. E., Elsabbagh, H. S., and El-Borai, N. B. 2021. Neuroprotective effect of sesamol against aluminum nanoparticle-induced toxicity in rats. Environmental Science and Pollution Research, 28,38, 53767–53780. https://doi.org/10.1007/s11356-021-14587-x
4. Alghriany, A. A. I., Omar, H. E. D. M., Mahmoud, A. M., and Atia, M. M. 2022. Assessment of the Toxicity of Aluminum Oxide and Its Nanoparticles in the Bone Marrow and Liver of Male Mice: Ameliorative Efficacy of Curcumin Nanoparticles. ACS Omega, 7,16, 13841–13852. https://doi.org/10.1021/acsomega.2c00195
5. Allain, C. C., Poon, L. S., Chan, C. S. G., Richmond, W., and Fu, P. C. 1974. Enzymatic Determination of Total Serum Cholesterol. Clinical Chemistry, 20,4, 470–475. https://doi.org/10.1093/clinchem/20.4.470
6. Azab, K. S., Mostafa, A. H. A., Ali, E. M. M., and Abdel-Aziz, M. A. S. 2011. Cinnamon extract ameliorates ionizing radiation-induced cellular injury in rats. Ecotoxicology and Environmental Safety, 74,8, 2324–2329. https://doi.org/10.1016/j.ecoenv.2011.06.016
7. Błaszczyk, N., Rosiak, A., and Kałużna-Czaplińska, J. 2021. The Potential Role of Cinnamon in Human Health. Forests, 12,5, 648. https://doi.org/10.3390/f12050648
8. Brancroft, J. ., and Gamble, M. 2008. Theory and Practice of Histological Techniques. Churchill and Livingstone; 2008, 31,5, 609–609. https://doi.org/10.1136/jcp.36.5.609-d
9. El-Borai, N. B., Elkhadrawey, B. A., AbuBakr, H. O., Anis, A., El-Bialy, B. E., Elsabbagh, H. S., and Abou-Zeid, S. M. 2022. Sesamol protects against aluminum oxide nanoparticles-induced hepatorenal toxicity in rats via modulation of oxidative stress, inflammation, apoptosis, and DNA damage. Environmental Toxicology, 37,8, 1914–1924. https://doi.org/10.1002/tox.23537
10. Elkomy, A., Aboubakr, M., Soliman, A., Abdeen, A., Abdelkader, A., and Hekal, H. 2016. Paracetamol induced hepatic toxicity and amelioration by cinnamon in rats. International Journal of Pharmacology and Toxicology, 4,2, 187. https://doi.org/10.14419/ijpt.v4i2.6529
11. Giera, M., Lingeman, H., and Niessen, W. M. A. 2012. Recent Advancements in the LC- and GC-Based Analysis of Malondialdehyde ,MDA,: A Brief Overview. Chromatographia, 75,9–10, 433–440. https://doi.org/10.1007/s10337-012-22371
12. Gojova, A., Guo, B., Kota, R. S., Rutledge, J. C., Kennedy, I. M., and Barakat, A. I. 2007. Induction of Inflammation in Vascular Endothelial Cells by Metal Oxide Nanoparticles: Effect of Particle Composition. Environmental Health Perspectives, 115,3, 403–409. https://doi.org/10.1289/ehp.8497
13. Harborne, J. B. 1973. Phytochemical Methods, A Guide to Modern Techniques of Plant Analysis. In London: Champon and Hall, Ltd. Springer Netherlands. https://doi.org/10.1007/978-94-009-5921-7
14. Huang, X.-J., Choi, Y.-K., Im, H.-S., Yarimaga, O., Yoon, E., and Kim, H.-S. 2006. Aspartate Aminotransferase (AST/GOT) and Alanine Aminotransferase (ALT/GPT), Detection Techniques. Sensors, 6,7, 756–782. https://doi.org/10.3390/s6070756
15. Hussain, S., Ashafaq, M., Alshahrani, S., Siddiqui, R., Ahmed, R. A., Khuwaja, G., and Islam, F. 2020. Cinnamon oil against acetaminophen-induced acute liver toxicity by attenuating inflammation, oxidative stress and apoptosis. Toxicology Reports, 7, 1296–1304. https://doi.org/10.1016/j.toxrep.2020.09.008
16. Hussain, Z., Khan, J. A., Arshad, M. I., Muhammad, F., and

Abbas, R. Z. 2021. Protective effects of cinnamon, cinnamaldehyde and kaempferol against acetaminophen-induced acute liver injury and apoptosis in mouse model. Pakistan Veterinary Journal, 41,1, 25–32. https://doi.org/10.29261/pakvetj/2020.090

17. Kiruthiga, P. V., Shafreen, R. B., Pandian, S. K., and Devi, K. P. 2007. Silymarin Protection against Major Reactive Oxygen Species Released by Environmental Toxins: Exogenous H 2 O 2 Exposure in Erythrocytes. Basic & Clinical Pharmacology & Toxicology, 100,6, 414–419. https://doi.org/10.1111/j.1742-7843.2007.00069.x
18. Korotkova, N. A., Baranovskaya, V. B., and Petrova, K. V. 2022. Microwave Digestion and ICP-MS Determination of Major and Trace Elements in Waste Sm-Co Magnets. Metals, 12,8, 1308. https://doi.org/10.3390/met12081308
19. Krause, B. C., Kriegel, F. L., Rosenkranz, D., Dreiack, N., Tentschert, J., Jungnickel, H., Jalili, P., Fessard, V., Laux, P., and Luch, A. 2020. Aluminum and aluminum oxide nanomaterials uptake after oral exposure - a comparative study. Scientific Reports, 10,1, 2698. https://doi.org/10.1038/s41598-020-59710-z
20. Li, H., Huang, T., Wang, Y., Pan, B., Zhang, L., Zhang, Q., and Niu, Q. 2020. Toxicity of Alumina Nanoparticles in The Immune System of Mice. Nanomedicine, 15,9, 927–946. https://doi.org/10.2217/nnm-2020-0009
21. Liu, H., Zhang, W., Fang, Y., Yang, H., Tian, L., Li, K., Lai, W., Bian, L., Lin, B., Liu, X., and Xi, Z. 2020. Neurotoxicity of aluminum oxide nanoparticles and their mechanistic role in dopaminergic neuron injury involving p53-related pathways. Journal of Hazardous Materials, 392,October 2019, 122312. https://doi.org/10.1016/j.jhazmat.2020.122312
22. M'rad, I., Jeljeli, M., Rihane, N., Hilber, P., Sakly, M., and Amara, S. 2018. Aluminium oxide nanoparticles compromise spatial learning and memory performance in rats. EXCLI Journal, 17, 200–210. https://doi.org/10.17179/excli2017-1050
23. Makwana, A. A., Fefar, D. T., Delvadiya, R. S., Thakore, K. A., Patel, S. S., Patel, U. D., Bhadaniya, A. R., and Gajera, K. G. 2024. Hepato-toxic effects of aluminium oxide nanoparticles following repeated dose 28-day oral exposure in Wistar rats. International Journal of Veterinary Sciences and Animal Husbandry, 9,2, 542–549. https://doi.org/10.22271/veterinary.2024.v9.i2h.1257
24. Morsy, G. M., Abou el-Ala, K. S., and Ali, A. A. 2016. Studies on fate and toxicity of nanoalumina in male albino rats: Oxidative stress in the brain, liver and kidney. Toxicology and Industrial Health, 32,2, 200–214. https://doi.org/10.1177/0748233713498462
25. Morsy, G. M., El-Ala, K. S. A., and Ali, A. A. 2016. Studies on fate and toxicity of nanoalumina in male albino rats. Toxicology and Industrial Health, 32,4, 634–655. https://doi.org/10.1177/0748233713504022
26. Nehru, B., and Anand, P. 2005. Oxidative damage following chronic aluminium exposure in adult and pup rat brains. Journal of Trace Elements in Medicine and Biology, 19,2–3, 203–208. https://doi.org/10.1016/j.jtemb.2005.09.004
27. Poborilova, Z., Opatrilova, R., and Babula, P. 2013. Toxicity of aluminium oxide nanoparticles demonstrated using a BY-2 plant cell suspension culture model. Environmental and Experimental Botany, 91, 1–11. https://doi.org/10.1016/j.envexpbot.2013.03.002
28. Rao, P. V., and Gan, S. H. 2014. Cinnamon: A Multifaceted Medicinal Plant. Evidence-Based Complementary and Alternative Medicine, 2014,1, 1–12. https://doi.org/10.1155/2014/642942
29. Rosalki, S. B., and Foo, A. Y. 1984. Two new methods for separating and quantifying bone and liver alkaline phosphatase isoenzymes in plasma. Clinical Chemistry, 30,7, 1182–1186. https://doi.org/10.1093/clinchem/30.7.1182
30. Sajid, M., Ilyas, M., Basheer, C., Tariq, M., Daud, M., Baig, N., and Shehzad, F. 2015. Impact of nanoparticles on human and environment: review of toxicity factors, exposures, control strategies, and future prospects. Environmental Science and Pollution Research, 22,6, 4122–4143. https://doi.org/10.1007/s11356-014-3994-1
31. Sakr, S. A., and Albarakai, A. Y. 2014. Effect of cinnamon on cypermethrin-induced nephrotoxicity in albino rats. International Journal of Advanced Research, 2,7, 578–586.
32. Sezgintürk, M. K., and Dinçkaya, E. 2011. Glutathione ,GSH, Determination by a Very Simple Electrochemical Method. International Journal of Peptide Research and Therapeutics, 17,2, 87–92. https://doi.org/10.1007/s10989-011-9243-2
33. Shrivastava, R., Raza, S., Yadav, A., Kushwaha, P., and Flora, S. J. S. 2014. Effects of sub-acute exposure to TiO 2 , ZnO and Al 2 O 3 nanoparticles on oxidative stress and histological changes in mouse liver and brain. Drug and Chemical Toxicology, 37,3, 336–347. https://doi.org/10.3109/01480545.2013.866134
34. Siddique, N. A., Mujeeb, M., Najmi, A. K., Aftab, A., and Aslam, J. 2011. Free radical scavenging and hepatoprotective activity of Aegle marmelos ,Linn, Corr leaves against carbon tetrachloride. International Journal Of Comprehensive Pharmacy, 02,08, 1–6.
35. Silva, V. S., Duarte, A. I., Rego, A. C., Oliveira, C. R., and Gonçalves, P. P. 2005. Effect of Chronic Exposure to Aluminium on Isoform Expression and Activity of Rat ,Na+/K+,ATPase. Toxicological Sciences, 88,2, 485–494. https://doi.org/10.1093/toxsci/kfi324
36. Stanley, J. K., Coleman, J. G., Weiss, C. A., and Steevens, J. A. 2010. Sediment toxicity and bioaccumulation of nano and micron-sized aluminum oxide. Environmental Toxicology and Chemistry, 29,2, 422–429. https://doi.org/10.1002/etc.52
37. Werner, E. 2003. Determination of Cellular H 2 O 2 Production. Science's STKE, 2003,168, 20773099. https://doi.org/10.1126/stke.2003.168.pl3
38. Yousef, M. I., Mutar, T. F., and Kamel, M. A. E.-N. 2019. Hepato-renal toxicity of oral sub-chronic exposure to aluminum oxide and/or zinc oxide nanoparticles in rats. Toxicology Reports, 6,April, 336–346. https://doi.org/10.1016/j.toxrep.2019.04.003
39. Yousef, M., Roychoudhury, S., Jafaar, K., Slama, P., Kesari, K., and Kamel, M. 2022. Aluminum Oxide and Zinc Oxide Induced Nanotoxicity in Rat Brain, Heart, and Lung. Physiological Research, 71,5, 677–694. https://doi.org/10.33549/physiolres.934831
40. Zhang, S., Bi, H., and Liu, C. 2007. Extraction of bio-active components from Rhodiola sachalinensis under ultrahigh hydrostatic pressure. Separation and Purification Technology, 57,2, 277–282. https://doi.org/10.1016/j.seppur.2007.04.022.