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# Amelioration Potency of a Nano-Therapeutic Drug in Rats with Uninephrectomy and Cisplatin-induced Toxicity

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**ABSTRACT**

While physicians describe drugs to treat diseases, these medications may have cytotoxic effects on certain organs, necessitating the use of some drugs to ameliorate such adverse effects. The study was conducted to investigate the protective behavior of nanoemulsified sodium salicylate on uninephrectomized rats injected with cisplatin to induce nephrotoxicity. Fifty adult male albino rats, aged five weeks and weighing approximately 100-120 g, were divided into five groups. The first group received 200 mg/kg/day i.p normal saline for 30 days. The second group was administrated 200 mg/kg/day of nanoemulsified salt of salicylic acid for 30 days. The third group, comprising uninephrectomized rats, was injected with two doses of cisplatin (20 mg/kg body weight) on alternate days from the start of the experiment to induce nephrotoxicity. The fourth group, also uninephrectomized, received 200 mg/kg/day i.p of nanoemulsified sodium salicylate for 30 days. The fifth group, uninephrectomized and treated with 200 mg/kg/day sodium salicylate nanoemlusion for 21 days, was subsequently injected with two doses of cisplatin, followed by continued nanoemulsified sodium salicylate treatment until day 30 from the start of the study. The results showed a significant increase in tissue inhibitor metalloproteinase 1 (TIMP-1), Hyaluronic acid (HA), malondialdehyde, kidney injury molecule -1(KIM-1), and nitric oxide in the nephrotoxic group injected with cisplatin compared to the control group. Additionally, there was an elevation in the mRNA expression of nephrotoxic group with uninephrectomy. However, nephrotoxic rats treated with nanoemulsified sodium salicylate exhibited only a modest increase in TIMP-1, HA, and KIM-1 levels, along with elevated expressions of podocin and nephrin compared to the healthy control group. These findings suggest that nanoemulsified sodium salicylate exerts a protective effect against cisplatin-induced nephrotoxicity in uninephrectomized.

**Keywords:** Cisplatin, Lateral nephrectomy, Nanoemlusion, Podocyte, Rat, Surgery

## INTRODUCTION

Drug toxicity is a major factor contributing to the withdrawal of medications from market. Between 1953 and 2013, over 400 medications were withdrawn, with 10% of causes related to neurotoxicity and 16% to cardiotoxicity (Onakpoya et al., 2016).

The considerable cost of drug development and research from 2009 to 2018 amounted to billions of dollars. A significant contributor to these costs is the high rate of failure during clinical trials and developmental studies (Wouters eta al., 2020; Rennane et al., 2021). Despite a 15.6% decline in sales in the pharmaceutical industry from 2008 to 2019, there was an increase in research and development intensity from 11.9% to 17.7%. Development and research spending for large pharmaceutical companies rose from 16.6% to 19.3%, while sales increased by 10.0% (from $380.0 to $418.0 billion) during the same period, even though the cost of drug development remained relatively stable or even decreased (Sertkaya et al., 2024). For instance, only about 15% of central nervous system drugs and 25% of cardiovascular drugs progress to phase 1 clinical trials (Amanat et al., 2022).

Antineoplastic drugs, widely used to treat various cancers, including ovarian, head, cervical, testicular carcinoma, and neck cancers, have demonstrated significant efficacy (Gómez-Sierra et al., 2018). Although the activity of antineoplastic drugs has been reported to be great, their use is often limited by severe side effects, such as hepatotoxicity,



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neurotoxicity, myelosuppression, ototoxicity, and nephrotoxicity (Pellacani and Eleftheriou, 2020). Several studies have revealed that cisplatin, a commonly used antineoplastic agent, accumulates in organs such as the liver and kidney (Palipoch and Punsawad, 2013; Apaydin et al., 2018; Aldossary, 2019).

Drug-induced toxicity, particularly nephrotoxicity, is a significant concern, leading to substantial health and economic losses (Frezza et al., 2010). The nephrotoxicity of cisplatin is attributed to its formation of highly active platinum complexes that bind to nucleophilic sites on DNA through an inter-strand and intra-strand crosslinking by nucleotide guanine. These symptoms result in denaturation and arrest cell cycle dewspite the mechanism of cisplatin nephrotoxicity change from its anticancer activity (Hanigan and Devarajan, 2003).

Cisplatin through cytochrome P450 mitochondrial dysfunction and microsome lead to the formation of reactive oxygen species (ROS) and damage to renal tissues. Elevated ROS levels contribute to acute failure of kidney through the induction of inflammatory cytokines, oxidative damage, tubule-interstitial inflammation, and necrosis of kidney tubular cells (Saisyo et al., 2016). Various adjuvants, including antioxidants, anti-apoptotic agents, and modulators of nitric oxide have been investigated for their potential protective effects against cisplatin-induced kidney injury (Ali and Al Moundhri, 2006).

Nasr (2014) demonstrated that cisplatin-induced hepatotoxicity in male experimental animals at a dose of 7.5 mg/kg i.p, altering lipid peroxidation and liver biomarkers, along with significant changes in antioxidant enzymes such as glutathione peroxidase and catalase. The formulation of nanoemulsified sodium salicylates, which offers a sustained effect due to its nanoformulation, has shown promise. Nanoemulsions based on silica nanoparticles are utilized for encapsulating sodium salicylates for several reasons, including the ability of nanoemulsified silica to carry bioactive ingredients and reduce their degradation. Encapsulating sodium salicylates move free to target through the bloodstream, induce disease targeting, and minimize the side effects on healthy organs.

This study explores the ameliorative effects of nanoemlusified sodium salicylates against nephrotoxicity induced by cisplatin in uninephrectomized experimental animals.

## MATERIALS AND METHODS

### Ethical approval

The study protocols and guide were approved by the National Research Centre Ethics Committee (Cairo, Egypt) under the registered ethical number 16/370.

### Chemical kits

Sodium salicylate, cisplatin, and TRIzol reagent were obtained from Sigma-Aldrich (St. Louis, MO, USA). Ultrapure water was prepared using a Millipore Milli-Q Plus system.

### Experimental animal

Fifty adult male Wister albino rats, five weeks old and weighing about 100-120 g, were procured from the Animal Housing Laboratory at the National Research Centre (NRC), Giza, Egypt, and were divided into five groups. The rats were housed under standard conditions, with a temperature of 25±2 °C and suitable ventilation. They were kept in qualified polypropylene cages with free access to standard diet and water.

### Experimental design

Following a 10-day acclimatization period, the fifty rats were randomly divided into five groups: The first (control) group received 200 mg/kg/day i.p normal saline for 30 days. The second (nanoemulsion) group was administrated 200 mg/kg/day of nanoemulsified salt of salicylic acid for 30 days. The third (nephrotoxic) group, including the uninephrectomized rats, were injected with two doses of cisplatin (20 mg/kg body weight) on alternate days from the start of the experiment to induce nephrotoxicity. The fourth (treatment) group, comprising uninephrectomized rats, were injected with 200 mg/kg/day i.p nano emulsion of sodium salicylate for 30 days. Finally, the fifth (pre-treatment and cisplatin) group, including uninephrectomized rats, were injected with 200 mg/kg/day sodium salicylate nanoemlusion for 21 days, followed by two doses of cisplatin (20 mg/kg, administered on alternate days). The nanoemulsified sodium salicylate treatment continued until the end of the 30-day study period.

### Surgical nephrectomy

Seven days before the experiment, the rats were anesthetized by inhalation of diethyl ether for 5 to 12 minutes, depending on their weight, with an ether volume of approximately 6 ml per hour. The surgical site was shaved, washed with soap and water, and sterilized with 70% alcohol followed by 4% tincture iodine. The incision in the muscles and

skin was done using a sterile cutter. The wound was widened till the kidney appeared using forceps. Separate ligation of the renal vein and renal artery was done with kidney excision. Lamberts sutures were used to close the wound.

**Sampling of kidney tissue and blood**

Thirty days after the start of the study, the rats were fasted for 12 hours. Blood samples were collected from the venous plexus of the eyes using a capillary tube with heparin. About 3.5 ml of blood was collected in tubes for biochemical analysis of the study's relevant biological parameters. One kidney was quickly removed from each rat, perfused, and rinsed with ice-cold saline to remove blood cells (Wu et al.,2017). The renal tissue was preserved in TRIzol reagent (Sigma-Aldrich, St. Louis, MO, USA) for gene expression analysis (Saleh et al., 2022).

**Investigation of biological markers**

The mRNA expression levels of various genes in renal tissues were quantified using real-time quantitative PCR (qRT-PCR). The serum levels of tissue inhibitor of metalloproteinases-1 (TIMP-1), hyaluronic acid (HA), and kidney injury molecule-1 (KIM-1) were measured in samples collected 30 days after the start of the experiment.

**Preparation of genes**

***Expression analysis of nephrotoxicity related genes***

***RNA isolation***

Total RNA was isolated from renal tissues of all experimental groups using TRIzol® reagent. The isolated RNA was then dissolved in deionized water. To eliminate any contaminating DNA, RNA samples were treated with RNAse-free DNAse. The purity of the RNA was assessed using spectrophotometry.

***Reverse transcription reaction***

First-strand cDNA synthesis was performed to generate cDNA copies from the isolated RNA. The reverse transcription reaction was carried out using the following program: about 600 seconds at room temperature, followed by 60 minutes at 42 °C, and 300 seconds at 95°C. The resulting cDNA was then collected in tubes and kept in tubes inside snow cups (Khalil and Booles, 2011).

***Quantitative Real Time-PCR***

Quantitative Real-Time PCR was conducted in a 0.025 mL reaction volume, which included 0.0125 mL of SYBR® Premix Ex TaqTM, 0.005 mL 0.002 mM antisense primer, 0.005 mL 0.002 mM sense primers, 0.0065 mL distilled water, and 0.005 mL of cDNA template. The qRT-PCR program consisted of the following steps: Step 1 involved an initial denaturation for 180 seconds at 95.0°C. Step 2 included 40 cycles. Step 3 involved generating a melting curve profile with about 71 turns, starting at 60°C and increasing by 0.5°C every 0.6 minutes until reaching 95.0°C (Khalil and Booles, 2011).

**Statistical investigations**

Statistical Analysis was carried out using one-way analysis of variance (ANOVA), followed by Bonferroni post hoc analysis. All data were expressed as mean ± standard error. The data were analyzed using SPSS software (Version 16). A p-value of less than 0.001 was considered statistically significant.

## RESULTS

Cisplatin-induced nephrotoxicity in the experimental animals led to a significant increase in serum levels of HA, TIMP-1, and KIM-1 ($p < 0.05$). However, in uninephrectomized animals treated with ameliorate, there was a significant but comparatively smaller elevation in the levels of HA, KIM-1, and TIMP-1 compared to healthy control animals ($p < 0.05$, Table 1).

**Table 1.** Effect of nano emulsified sodium salicylate on adult male albino rats weighing about 100-120g aged 5 weeks

| Treatments | HA (Pg\ml) | TIMP-1 (Pg\ml) | KIM-1 (Pg\ml) |
|---|---|---|---|
| Control | 40.5 ± 1.07 | 68 ± 1.04 | 24 ± 1.18 |
| Carrier | 42.9 ± 2.02 | 78 ± 1.06 | 25 ± 1.13 |
| CisP | 90.9 [a,b,d] ± 3. 5 | 128[a,b,d] ± 2.16 | 158 [a,b,d] ± 1.18 |
| SiNPs@sod | 43.6 [c,,e] ±1.17 | 86[c,,e] ± 1.04 | 99 [c,e] ±1.19 |
| SiNPs@sod-CisP | 49.4[a,b,c] ± 3.6 | 77 [a,b,c] ± 1.07 | 53 [a,b,c] ± .99 |

*Results are expressed as means ± SD for 10 animals in certain 5 sets. [a] all results in comparison with the normal group, [b]all result in comparison to SiNPs group. [c] Significant difference compared to the Cisplatin group. [d] Significant difference compared to SiNPs@sod group [e] Significant difference compared to SiNPs@sod- Cisplatin group significant at $P < 0.05$. The superscript (a,b,c,d) shows a significant difference at $p < 0.05$. **HA: Hyaluronic acid; TIMP -1: Tissue inhibitor metalloproteinase 1; KIM -1: kidney injury molecule -1, First group: Control, second group: Nanoemulsion, third (nephrotoxic) group: Injected with two doses of cisplatin. Fourth (treatment) group: Uninephrectomized rats, were injected with 200 mg/kg/day i.p nanoemulsion of sodium salicylate. Fifth (pre-treatment and cisplatin) group: Uninephrectomized rats, injected with 200 mg/kg/day sodium salicylate nanoemlusion, followed by two doses of cisplatin.

**Oxidative stress markers in kidney homogenate**

Kidney malondialdhyde and nitric oxide levels were significantly elevated by 116% and 128%, respectively, in the Cisplatin group compared to the control, indicating increased oxidative stress. However, both renal malondialdhyde and nitric oxide levels showed a significant decrease ($p < 0.05$) in the group treated with nano-emulsified sodium salicylate compared to the normal ($p < 0.05$ ) Conversely, rats treated with sodium salicylate alone or with the nanoemulsified form exhibited no significant changes in oxidative stress markers (Table 2).

**Nephrin and Podocin genes**

The expression levels of podocin and nephrin showed a significant decrease in the Cisplatin-only group with a 1.2- and 1.3-fold reduction, respectively ($p < 0.05$). However, uni nephrectomized animals treated with nano emulsified exhibited only a modest reduction in the expression levels of nephrin and podocin genes with a 0.2-fold decrease compared to healthy animals. In contrast, rats treated with either silicate or the nanoemulsified form of salicylic acid showed no significant variation in gene expression levels compared to the normal control group ($p < 0.05$) (Table 3).

**Table 2.** Effects of nano emulsified sodium salicylate on oxidative stress indicators of adult male albino rats weighing about 100-120g aged 5 weeks

| Treatments | MD (nmol/l) | No(nmol/l) |
|---|---|---|
| Control | 90.9 ± 5.2 | 10 ± 0.45 |
| Carrier | 91.57 ± 2.8 | 9.1 ± 0.3 |
| CisP | $190.1^{a,b,d,e}$ ± 12.5 | $50.2^{a,b,d,e}$ ± 0.69 |
| SiNPs@sod | $100.6^{c,e}$ ± 5.04 | $12.02^{c,e}$ ± 0.42 |
| SiNPs@sod-CisP | $105.2^{a,b}$ ± 4.1 | $19.9 \pm 0.79^{a,b}$ |

*Results are revealed as means ± SD for 10 animals in certain 5 sets. The superscript ($^{a,b,c,d}$) shows a significant difference at $p < 0.05$. ** MD: Malondialdehyde; NO: Nitric oxide; CisP: Cisplatin; SiNPs@sod: Sodium salicylate nanoparticles; SiNPs@sod-CisP: Sodium salicylate nanoparticle with cisplatin, first group: Control, second group: Nanoemulsion, third (nephrotoxic) group: Injected with two doses of cisplatin. Fourth (treatment) group: Uninephrectomized rats, were injected with 200 mg/kg/day i.p nano emulsion of sodium salicylate. Fifth (pre-treatment and cisplatin) group: Uninephrectomized rats, injected with 200 mg/kg/day sodium salicylate nanoemlusion, followed by two doses of cisplatin.

**Table 3.** Effect of nano emulsified sodium salicylate 0n expression of Podocin and Nephrin genes in adult male albino rats weighing 100 -120 g aged 5 weeks

| Treatments | Nephrin | Podocin |
|---|---|---|
| Control | 1.74 ± 0.012 | 2.1 ± 0.03 |
| Carrier | 1.8 ± 0.004 | 2.2 ± 0.008 |
| CisP | $0.9^{abde}$ ± 0.002 | $0.76^{abde}$ ± 0.004 |
| SiNPs@sod | 1.07 ± 0. 14 | 1.9 ± 0.012 |
| SiNPs@sod-CisP | $1.16^{ab}$ ± 0.017 | $1.5^{ab}$ ± 0.016 |

*Values are represented as means ± SD for 10 rats in each group and 5 sets. The superscript ($^{a,b,c,d}$) shows a significant difference at $p < 0.05$. CisP: Cisplatin; SiNPs@sod: Sodium salicylate nanoparticles; SiNPs@sod-CisP: Sodium salicylate nanoparticle with cisplatin, first group: Control, second group: Nanoemulsion, third (nephrotoxic) group: Injected with two doses of cisplatin. Fourth (treatment) group: Uninephrectomized rats, were injected with 200 mg/kg/day i.p nano emulsion of sodium salicylate. Fifth (pre-treatment and cisplatin) group: Uninephrectomized rats, injected with 200 mg/kg/day sodium salicylate nanoemlusion, followed by two doses of cisplatin.

## DISCUSSION

Many systemic approaches with selective protection of normal cells from the toxic effects of chemotherapy also have been investigated (Brizel, 1998). The current study investigated the nephro-protective potentials using the technique of nanotechnology drug delivery as a cytoprotector. According to the obtained results, cisplatin injection significantly increased the serum HA. The nephrotoxic effect of cisplatin on serum HA was previously reported (Akin et al., 2017). Extracellular matrix markers undergo both qualitative and quantitative alterations during the process of fibrosis. Potential markers of fibrosis include glycosaminoglycans and glycoproteins found in the extracellular matrix, as well as the synthesis of matrix and enzymes associated with degradation, along with collagen synthesis and the enzymes involved in its breakdown. One of the direct markers of fibrosis is HA, which is found to be elevated in chronic kidney disease. Results in the present study revealed that the cisplatin group recorded a significant increase in serum TIMP-1 compared to control. Additionally, serum TIMP- 1 mean value was significantly increased in the cisplatin group compared to the carrier group. The result is in agreement with Tianhui et al. (2012), who suggested that the activation of TIMP1 is responsible for the deposition of extracellular matrix following kidney damage. Protection of nephrotoxic rats with

nanoemlusified salt of salicylic acid resulted in a deceased TIMP-1 relative nonprotected group, however, it was still higher than its matched value in the control group. The ameloriting effect of encapsulated sodium salycsilate on serum TIMP-1 was reported previously (Cheng et al., 2020). In this work increased serum KIM-1 in the cisplatin group was noted and agrees with Takaharu et al., (2004). Takaharu et al., (2004) reported that KIM-1 may be a sensitive general renal injury or early repair biomarker in animals exposed to nephrotoxicants. Measurement of KIM-1 protein expression may be beneficial for the detection of abnormalities of tubular epithelial cells during injury/repair in the kidney. KIM-1 may be useful in preclinical and clinical studies vital to drug development and evaluation. It may also serve in the monitoring of disease states that manifest as injury to the proximal tubule and be useful in guiding interventional strategies (Tanase et al., 2019; Chalo and Aydilek, 2022). Administration of nano-emulsified sodium salicylate ameliorated Cisplatin-induced changes in multiple blood and urine variance, contributing to a notable recovery. Results of the present study revealed no significant change in kidney Malondialdhyde and nitric oxide in the carrier group compared to the control. Furthermore, a significant increase in kidney MDA and NO in the cisplatin cisplatin-injected group was observed compared to the control and carrier groups. These results are in agreement with Naqshbandi et al., (2013). Oxidative stress plays a pivotal role in Cisplatin-induced toxicity, leading to various changes in blood and urine parameters. Under normal physiological conditions, the generation and elimination of ROS in tissues are balanced by endogenous scavenging systems such as catalases, reduced glutathione, and superoxide dismutase. Nanoemulsified sodium salicylate appears to mimic these mechanisms. Elevated ROS levels damage multiple macromolecules, including DNA, lipids, and proteins. Oxygen radicals play a role in the nephrotoxicity enhanced by cisplatin lead to damage of macromolecules in cells. Elevating oxidative stress causes necrosis and cellular injury in the liver, kidney, and other tissues (Naqshbandi et al., 2013). Antioxidant enzymes, such as paraoxinase scavenge free radicals or convert them into non-toxic forms (Zheng et al., 2008).

The results of the present study showed that cisplatin injection elevated the levels of lipid peroxidation markers, such as nitric oxide and malondialdhyde (Ognjanović et al., 2012; Koyuncu et al., 2017). In contrast, nano emulsified sodium salicylate decreased lipid peroxidation levels in the kidney and helped retain normalized antioxidant enzyme levels. Patients with chronic renal failure exhibited multiple changes in amino acid and protein metabolism, including increased levels of homocysteine amino acid (Guldener, 2006). Elevated homocysteine levels are frequently observed in individuals suffering from renal failure, with multiple studies establishing a correlation between heightened homocysteine levels and a significant reduction in glomerular filtration rate (Ninomiya et al., 2004). These observations align with the results of the present study, where homocysteine levels increased in the cisplatin toxic group compared to the healthy group. Acute kidney damage increases homocystein due to alterations in the disposal of homocystein or impaired metabolism (Guldener, 2006). The enhancement observed with nano-emlusified salt of salicylic acid in the present study could reduce the cisplatin-induced elevation of TIMP1 of Cis- c, indicating the significant protective role of this nano-formulated medication in preventing renal injury (Al Nahyah, 2021). The TIMP-1 is responsible for the precipitation of the extracellular matrix (Cong et al., 2013).

The filtration of blood components via the formation of urine in the glomerulus capsule is one of the key functions of the kidney. The filtration pores between adjacent foot processes are crucial for maintaining the integrity of the glomerular filter (Murray and Paolini, 2023). In experimental animals, several studies have highlighted the direct role of the slit diaphragm in the progression of proteinuria, suggesting its importance in preserving the glomerular filtration barrier (Kawachi and Shimizu, 2000; Kocylowski et al., 2022).

The current study indicated that the level of tissue nephrin mRNA and podocin mRNA significantly decreased in the cisplatin group compared to the normal control and carrier groups, which was previously reported by Kaveripakam and Adikay (2017). Podocin and nephrin are essential proteins located in the podocyte slit diaphragm, significantly contributing to the functionality of the glomerular filter (Huber et al., 2003). Researchers indicated the downregulation of nephrin mRNA in the cisplatin group, including the characterization of structural alteration in glomerular epithelial cells, alteration of foot processes, and subsequent filtration slits, as well as the loss of podocytes. From the current perspective and alongside the results of the present study, the loss of podocytes is closely related to progressive glomerular injury or both. Protection with carrier recorded upregulation of nephrin mRNA compared to the cisplatin group, indicating an improvement in kidney functions (Wang et al., 2002).

## CONCLUSION

The use of unsaturated lipid and polar solvents in the formulation of nanoemlusified sodium salicylate as a drug delivery system has shown significant potential in improving kidney disorders. These findings suggest that nanoemulsified sodium salicylate exerts a protective effect against cisplatin-induced nephrotoxicity in uninephrectomized. Further studies are required to evaluate the effects of nano-emulsified sodium salicylate on the toxicity of other internal organs.

## DECLARATIONS

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**Availability of data and materials**

Raw data or additional supporting information is available upon request from the corresponding author.

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**Authors' contributions**

This study was completed with the assistance and participation of all authors. The methods and design of the study (especially laboratory works) were developed by Rehab A. Mohamed, Nadia A. Mohamed, and Hend M. Ahmed. The drafting, analysis and proofreading were conducted by Rehab A. Mohamed, Luneva Albina Vladimirovna, and Lysenko Yuri Andreevich. All authors checked and approved the final edition of the manuscript before publication.

**Competing interests**

The authors do not have any conflict of interest.

**Ethical considerations**

The article was checked for plagiarism during the writing of the draft of the manuscript and also the final edition was originally prepared for submission. The authors confirm that all authors have reviewed and submitted the original manuscript to this journal for the first time.

## REFERENCES

Akin D, Ozmen S, Yilmaz ME. Hyaluronic Acid as a New Biomarker to Differentiate Acute Kidney Injury from Chronic Kidney Disease. Iranian journal of kidney diseases,11(6):409-413. Available at: https://pubmed.ncbi.nlm.nih.gov/29190600/

Al Nahyah KS (2021). Utilization of nanotechnology and hepatoprotective agents to improve the bioavailability and to reduce the toxicity associated with isotretinoin. Doctoral Dissertation, King Abdulaziz University, Saudi Arabia. Available at: https://platform.almanhal.com/Files/4/235749

Aldossary SA (2019). Review on pharmacology of cisplatin: Clinical use, toxicity and mechanism of resistance of cisplatin. Biomedical and Pharmacology Journal, 12(1): 7-15. DOI: https://www.doi.org/10.13005/bpj/1608

Ali BH and Al Moundhri MS (2006). Agents ameliorating or augmenting the nephrotoxicity of cisplatin and other platinum compounds: A review of some recent research. Food and Chemical Toxicology, 44(8): 1173-1183. DOI: https://www.doi.org/10.1016/j.fct.2006.01.013

Amanat M, Nemeth CL, Fine AS, Leung DG, and Fatemi A (2022). Antisense oligonucleotide therapy for the nervous system: From bench to bedside with emphasis on pediatric neurology. Pharmaceutics, 14(11): 2389. DOI: https://www.doi.org/10.3390/pharmaceutics14112389

Apaydin FG, Kaltalıoğlu K, Balabanlı B, and Cevher ŞC (2018). Morin and hesperidin ameliorate cisplatin-induced hepatotoxicity and nephrotoxicity in rats: A histopathological study. Gazi University Journal of Science, 31(2): 399-406. Available at: https://dergipark.org.tr/en/pub/gujs/issue/37206/337235

Brizel DM (1998). Future directions in toxicity prevention. In Seminars in Radiation Oncology, 8 (Suppl 1): 17-20. PMID: 9794997.

Chalo S, and Aydilek İB (2022). A New Preprocessing Method for Diabetes and Biomedical Data Classification. Qubahan Academic Journal, 2(4):6-18. DOI: https://www.doi.org/10.48161/qaj.v2n4a135

Cheng H, Lv M, Mi R, and Xue G (2020). Amifostine ameliorates cerebral ischaemia-reperfusion injury via p38-mediated oxidative stress and mitochondrial dysfunction. Folia neuropathologica, 58(4): 334-346. DOI: https://www.doi.org/10.5114/fn.2020.102436

Cong M, Liu T, Wang P, Fan X, Yang A, Bai Y, Peng Z, Wu P, Xiaofei Tong, Jing Chen et al. (2013). Antifibrotic effects of a recombinant adeno-associated virus carrying small interfering RNA targeting TIMP-1 in rat liver fibrosis. The American Journal of Pathology, 182(5): 1607-1616. DOI: https://www.doi.org/10.1016/j.ajpath.2013.01.036

Frezza M, Hindo S, Chen D, Davenport A, Schmitt S, Tomco D, and Ping Dou Q (2010). Novel metals and metal complexes as platforms for cancer therapy. Current Pharmaceutical Design, 16(16): 1813-1825. DOI: https://www.doi.org/10.2174/138161210791209009

Gómez-Sierra T, Eugenio-Pérez D, Sánchez-Chinchillas A, and Pedraza-Chaverri J (2018). Role of food-derived antioxidants against cisplatin induced-nephrotoxicity. Food and Chemical Toxicology, 120: 230-242. DOI: https://www.doi.org/10.1016/j.fct.2018.07.018

Guldener C (2006). Why is homocysteine elevated in renal failure and what can be expected from homocysteine-lowering?. Nephrology Dialysis Transplantation, 21(5): 1161-1166. DOI: https://www.doi.org/10.1093/ndt/gfl044

Hanigan MH and Devarajan P (2003). Cisplatin nephrotoxicity: Molecular mechanisms. Cancer Therapy, 1: 47. Available at: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2180401/

Huber TB, Simons M, Hartleben B, Sernetz L, Schmidts M, Gundlach E, Saleem MA, Walz G, Benzing T (2003). Molecular basis of the functional podocin-nephrin complex: Mutations in the NPHS2 gene disrupt nephrin targeting to lipid raft microdomains. Human Molecular Genetic, 12(24): 3397-3405. DOI: https://www.doi.org/10.1093/hmg/ddg360

Kaveripakam S and Adikay S (2017). Amelioration of Cisplatin- Induced Nephrotoxicityby Roots of Anthocephalus cadamba. Biomedical & Pharmacology Journal; 10(3): 1433-1439. DOI: https://www.doi.org/10.13005/bpj/1250

Khalil W and Booles H (2011). Protective role of selenium against over-expression of cancer-related apoptotic genes induced by-cresol in rats. Archives of Industrial Hygiene and Toxicology, 62(2): 121-129. DOI: https://www.doi.org/10.2478/10004-1254-62-2011-2074

Kawachi H, and Shimizu F (2000). Molecular composition and function of the slit diaphragm: nephrin, the molecule responsible for proteinuria. Clinical and experimental nephrology, 4:161-72. DOI: https://www.doi.org/10.1007/s101570070017

Kocylowski MK, Aypek H, Bildl W, Helmstädter M, Trachte P, Dumoulin B, and et al. (2022). A slit-diaphragm-associated protein network for dynamic control of renal filtration. Nature Communications, 13(1):6446. Available at: https://www.nature.com/articles/s41467-022-33748-1

Koyuncu I, Kocyigit A, Gonel A, Arslan E, and Durgun M (2017). The protective effect of Naringenin-oxime on cisplatin-induced toxicity in rats. Biochemistry Research International, 2017: 9478958. DOI: https://www.doi.org/10.1155/2017/9478958

Mondi S, Kvsrg P, Jhansi D, Vijay R, and Rao VU (2014). Prophylactic and curative effect of ethanolic extract of Bassiamalabaricabark against cisplatin induced nephrotoxicity. Asian Journal of Pharmaceutical and Clinical Research, 7(4): 143-146. Available at: https://journals.innovareacademics.in/index.php/ajpcr/article/view/1205

Murray IV, and Paolini MA (2023). Histology, kidney and glomerulus. StatPearls [Internet]. Medline. Available at: https://pubmed.ncbi.nlm.nih.gov/32119431/

Naqshbandi A, Rizwan S, and Khan F (2013). Dietary supplementation of flaxseed oil ameliorates the effect of cisplatin on rat kidney. Journal of Functional Foods, 5(1): 316-326. DOI: https://www.doi.org/10.1016/j.jff.2012.11.002

Nasr AY (2014). Aged garlic extract protects against cisplatin-induced hepatotoxicity in adult male rats: biochemical and ultrastructure study. Life Science Journal, 11(12): 92-101. Available at: http://www.lifesciencesite.com/lsj/life1112/016_26087life111214_92_101.pdf

Naziroğlu M, Karaoğlu A, and Aksoy AO (2004). Selenium and high dose vitamin E administration protects cisplatin-induced oxidative damage to renal, liver and lens tissues in rats. Toxicology, 195(2-3): 221-230. DOI: https://www.doi.org/10.1016/j.tox.2003.10.012

Ninomiya T, Kiyohara Y, Kubo M, Tanizaki Y, Tanaka K, Okubo K, Nakamura H, Hata J, Oishi Y, Kato I et al. Hyperhomocysteinemia and the development of chronic kidney disease in a general population: The Hisayama study. American Journal of Kidney Diseases, 44(3): 437-445. DOI: https://www.doi.org/10.1053/j.ajkd.2004.05.024

Ognjanović BI, Djordjević NZ, Matić MM, Obradović JM, Mladenović JM, Štajn AŠ, and Saičić ZS (2012). Lipid peroxidative damage on cisplatin exposure and alterations in antioxidant defense system in rat kidneys: A possible protective effect of selenium. International Journal of Molecular Sciences, 13(2): 1790-1803. DOI: https://www.doi.org/10.3390/ijms13021790

Onakpoya IJ, Heneghan CJ, and Aronson JK (2016). Post-marketing withdrawal of 462 medicinal products because of adverse drug reactions: a systematic review of the world literature. BMC Medicine, 14: 10. DOI: https://www.doi.org/10.1186/s12916-016-0553-2

Palipoch S and Punsawad C (2013). Biochemical and histological study of rat liver and kidney injury induced by cisplatin. Journal of Toxicologic Pathology, 26(3): 293-299. DOI: https://www.doi.org/10.1293/tox.26.293

Pellacani C and Eleftheriou G (2020). Neurotoxicity of antineoplastic drugs: Mechanisms, susceptibility, and neuroprotective strategies. Advances in Medical Sciences, 65(2): 265-85. DOI: https://www.doi.org/10.1016/j.advms.2020.04.001

Rennane S, Baker L, and Mulcahy A (2021). Estimating the cost of industry investment in drug research and development: a review of methods and results. INQUIRY: The Journal of Health Care Organization, Provision, and Financing, 58: 1-11. DOI: https://www.doi.org/10.1177/00469580211059731

Saisyo A, Nakamura H, Fang J, Tsukigawa K, Greish K, Furukawa H, and Maeda H (2016). pH-sensitive polymeric cisplatin-ion complex with styrene-maleic acid copolymer exhibits tumor-selective drug delivery and antitumor activity as a result of the enhanced permeability and retention effect. Colloids and Surfaces B: Biointerfaces, 138: 128-137. DOI: https://www.doi.org/10.1016/j.colsurfb.2015.11.032

Saleh H, Salama M, and Hussein RM (2022). Polyethylene glycol capped gold nanoparticles ameliorate renal ischemia–reperfusion injury in diabetic mice through AMPK-Nrf2 signaling pathway. Environmental Science and Pollution Research, 29(51):77884-907. DOI: https://www.doi.org/10.1007/s11356-022-21235-5

Sertkaya A, Beleche T, Jessup A, and Sommers BD (2024). Costs of drug development and research and development intensity in the US, 2000-2018. JAMA Net Open, 7(6): e2415445. DOI: https://www.doi.org/10.1001%2Fjamanetworkopen.2024.15445

Takaharu I, Hung CC, Yang SA, Stevens JL, Bonventre JV. Kidney injury molecule-1: a tissue and urinary biomarker for nephrotoxicant-induced renal injury. American Journal of Physiology-Renal Physiology, 286(3): F552-F563. Available at: https://journals.physiology.org/doi/full/10.1152/ajprenal.00285.2002

Tanase DM, Gosav EM, Radu S, Costea CF, Ciocoiu M, Carauleanu A, and et al. (2019). The predictive role of the biomarker kidney molecule-1 (KIM-1) in acute kidney injury (AKI) cisplatin-induced nephrotoxicity. International journal of molecular sciences, 20(20): 5238. DOI: https://www.doi.org/10.3390/ijms20205238

Wang SX, Rastaldi MP, Pätäri A, Ahola H, Heikkilä E, and Holthöfer H (2002). Patterns of nephrin and a new proteinuria-associated protein expression in human renal diseases. Kidney International, 61(1): 141-147. DOI: https://www.doi.org/10.1046/j.1523-1755.2002.00114.x

Wouters OJ, McKee M, and Luyten J (2020). Estimated Research and Development Investment Needed to Bring a New Medicine to Market, 2009-2018. JAMA, 323(9): 844-853. Available at: https://jamanetwork.com/journals/jama/article-abstract/2762311

Wu J, Pan X, Fu H, Zheng, Dai Y, Yin Y, Chen Q, Hao Q, Bao D, and Hou D (2017). Effect of curcumin on glycerol-induced acute kidney injury in rats. Scientific Reports, 7: 10114. DOI: https://www.doi.org/10.1038/s41598-017-10693-4

Zheng W, Huang LZ, Zhao L, Wang B, Xu HB, and Wang GY (2008). Superoxide dismutase activity and malondialdehyde level in plasma and morphological evaluation of acute severe hemorrhagic shock in rats. The American Journal of Emergency Medicine, 26(1): 54-58. DOI: https://www.doi.org/10.1016/j.ajem.2007.02.007