www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.03
TPI 2021; 10(2): 567-571

www.thepharmajournal.com
Received: 08-12-2020
Accepted: 22-01-2021

K Sneha
P.G Scholar, Department of Veterinary Pharmacology and Toxicology, College of Veterinary Science, Rajendranagar, Hyderabad, Telangana, India

A Gopala Reddy
Professor and University Head, Department of Veterinary Pharmacology and Toxicology and Registrar of PVNRTVU, College of Veterinary Science, Rajendranagar, Hyderabad, Telangana, India

M Usha Rani
Professor and Head, Department of Veterinary Pharmacology and Toxicology, College of Veterinary Science, Rajendranagar, Hyderabad, Andhra Pradesh, India

B Ramya
Assistant Professor, Veterinary Clinical Complex, College of Veterinary Science, Rajendranagar, Hyderabad, Andhra Pradesh, India

P Shiva Kumar
Assistant Professor, Department of Veterinary Pharmacology and Toxicology, College of Veterinary Science, Rajendranagar, Hyderabad, Andhra Pradesh, India

B Anil Kumar
Assistant Professor, Department of Veterinary Pharmacology and Toxicology, College of Veterinary Science, Rajendranagar, Hyderabad, Andhra Pradesh, India

Corresponding Author:
K Sneha
P.G Scholar, Department of Veterinary Pharmacology and Toxicology, College of Veterinary Science, Rajendranagar, Hyderabad, Telangana, India

Amelioration of experimental hepatotoxicity and nephrotoxicity due to cisplatin by *Terminalia arjuna* in comparison to taurine

K Sneha, A Gopala Reddy, M Usha Rani, B Ramya, P Shiva Kumar and B Anil Kumar

Abstract
The ameliorative potential of aqueous leaf extract of *Terminalia Arjuna* and taurine were studied against Cisplatin induced nephrotoxicity and hepatotoxicity. Total 36 male *Wistar* rats were separated into six groups (n = 6). Group 1 was maintained as normal control. A single dose of Cisplatin (5 mg/kg i.p) was administered on day 1 to groups 2, 5 and 6. Groups 3 and 5 were administered for 14 days (p/o) with aqueous leaf extract of *Terminalia Arjuna* @ 400 mg/kg bodyweight from day 1. Group 4 and 6 received received taurine @ 1000 mg/kg body weight orally for 14 days. After the experimental procedure, body weight, total protein and prothrombin time were assessed.
In the present study, body weights and total protein showed a significant ($P < 0.05$) reduction in group 2 when compared with group 1 and there was significant ($P < 0.05$) improvement in the treatment groups 5 and 6. The results of prothrombin time revealed a significant ($P < 0.05$) increase in group 2 and significant ($P < 0.05$) decrease in groups 5 and 6 compared to group 2. In conclusion, aqueous leaf extract of *Terminalia Arjuna* and taurine were found to possess protective action against Cisplatin induced nephrotoxicity and hepatotoxicity.

Keywords: Cisplatin, *Terminalia Arjuna*, taurine, nephrotoxicity and hepatotoxicity

Introduction
Cisplatin is inorganic platinum based chemotherapeutic drug[1] and it has been shown to be effective against different tumor types includes head, neck, lung, esophageal, stomach, urine bladder, skin prostate, lymphoma and neuroblastoma, myeloma, sarcoma, mesothelima, cervical and osteosarcoma [2]. Cisplatin reacts with nucleophilic sites in DNA and causes damage to tumors via apoptosis by activating some intracellular signaling pathways [3]. Despite its valuable antitumor action, Cisplatin has various adverse effects, which influence the gastrointestinal, neurological, and hematological system [4]. Its administration has been accompanied by nephrotoxicity [5], hepatotoxicity [6] and cardiotoxicity[7]. Different mechanisms have also been reported describing the Cisplatin toxicity. Oxidative stress plays an important role in Cisplatin toxicity [8], adverse effects were due to increased production of reactive oxygen species [9] and also induces lipid peroxidation, inflammation and hypoxia [10]. These injuries reduce glomerular infiltration and cause acute nephrotoxicity [11]. Also, hepatotoxicity is associated to oxidative damage and mitochondrial dysfunction [12].
Hence there is a necessity for investigation of ways to prevent the toxicity of Cisplatin treatment. Herbal medicines can be used to prevent and treat the diseases [13]. *Terminalia Arjuna*, commonly known as Arjuna belongs to family Combretaceae. It contains several bioactive compounds which provide various health benefits, such as many specific phytoconstituents including arjunolic acid, triterpene glycosides-arjunetin, arjunoglucoside, falvonoids and glycosides etc., [14]. Clinical evaluation of various preparations from *Terminalia Arjuna* demonstrated its beneficial role in the treatment of coronary artery disease. In addition, the *Terminalia arjuna* bark powder also possess diverse beneficial properties, such as anti-carcinogenic in human and mice [15], anti-dyslipidaemic in rat, anti hypocholesterolaemia in rabbit, anti-inflammatory and antioxidant in rat [16]. Taurine (2-aminoethane sulfonic acid) is the major intracellular free ß-amino acid, it acts as an antioxidant in a variety of *invitro* and *invivo* systems. It is present in many food stuffs, such as milk, meats and seafood and also found in high intracellular concentrations in most animal tissue [17]. Taurine can protect various organs from toxin and drug-induced pathophysiology [18].

Besides, its mechanism of action as a protective agent has also been established in many organ dysfunctions [19, 20]. Therefore, in the present study, we aimed to investigate whether taurine could offer any protection against Cisplatin-induced toxicity using rat as the animal model.

Material and methods

Chemicals

All chemicals were of analytical grade and they are obtained from Qualigens Pvt. Ltd., Mumbai and SRL Pvt. Ltd., Mumbai, India.

Plant material and preparation of leaf extract

The fresh leaves of *T.arjuna* plant were collected from Hyderabad, India. The plant species were authenticated by Scientist, Agricultural College, Hyderabad, India. The fresh leaves of *T.arjuna* were washed twice with distilled water and shade dried at room temperature for 40-45 days. Leaves were powdered using a mechanical blender. Thereafter, 1 g powder was mixed with 100 ml of boiled distilled water and stirred on hot plate for 15 minutes. After the process, extract was filtered through Whatman No. 1 filter paper. The filtrate was kept at low temperature (4 °C) for further use [21].

Animals and Experimental Design

Thirty six male *Wistar* rats aged about 3 months with an average body weight of 180 ± 10 g were obtained from Vyas labs, Hyderabad. They are divided into six equal groups (n=6) with different treatments (Table 1). The animals housed in polypropylene cages, under controlled environmental conditions (20–22ºC) and 12 hour dark and light cycles with sterilized dried clean, autoclaved rice husk was used as bedding material, which was changed on alternate days. The rats were maintained on standard balanced diet with drinking water *ad libitum* throughout the experimental period. All the experimental procedures and protocols used in this study were reviewed and approved by the Institutional Animal Ethical Committee (No.5/22/C.V.Sc., Hyd. IAEC- Rats/29.02.2020) and were in accordance with the guidelines of the Committee for the purpose of Control and Supervision of Experiments on Animals (CPCSEA).

Table 1: Experimental design with group wise treatment protocol

Groups	Treatments	No. of Animals
1.	Normal saline for 14 days (Control)	6
2.	Cisplatin Control (5mg/kg B.wt. i.p single dose on day 1)	6
3.	Aqueous leaf extract of *Terminalia arjuna* Control (400mg/kg B.wt Oral Route for 14 days)	6
4.	Taurine Control (1000mg/kg B.wt Oral Route for 14 days)	6
5.	Cisplatin (single dose on day 1) + Aqueous leaf extract of *Terminalia arjuna*(1-14 days)	6
6.	Cisplatin (single dose on day 1) + Taurine (1-14 days)	6

Blood collection After completion of 14 days, the blood samples were collected from retro-orbital plexus of experimental rats for further investigation.

Analysis

Rat body weights were taken on 7th and 14th day. Total protein is analyzed by kits from ERBA diagnostics Ltd, Surat, India. Prothrombin time was estimated based on time required for clot formation after adding thromboplastin reagent to normal anticoagulated plasma.

Statistical Analysis

Data were subjected to statistical analysis by applying one-way analysis of variance (ANOVA) using the statistical package for social sciences (SPSS). Differences between means were tested using Duncan's multiple comparison tests and significance was set at $P< 0.05$.

Results

The various experimental investigations recorded were group wise mean body weight, total protein and prothrombin time.

Body weight changes

Significant reduction ($P < 0.05$) in body weight (g) was observed in Cisplatin treated rats (group 2) as compared with non-toxic control (group 1, 3 and 4). Rats treated with aqueous leaf extract of *Terminalia arjuna* (group 5) and taurine (group 6) showed significant increase ($P < 0.05$) in body weight when compared with group 2 (Table 2 and Figure 1).

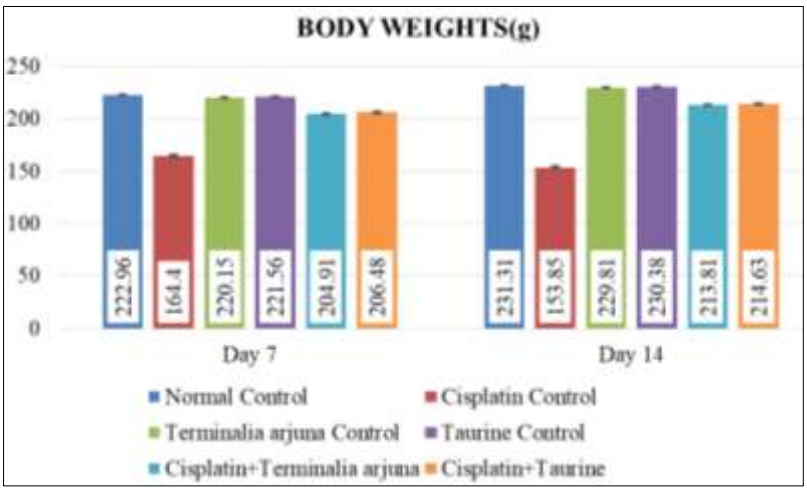

Fig 1: Average body weights (g) of different groups of rats

Total protein

Total protein (g/dl) level found to be significantly decreased ($P < 0.05$) in rats treated with only Cisplatin (group 2), whereas treatment with the aqueous leaf extract of *Terminalia arjuna* (group 5) and taurine (group 6) was found to protect the rats from such effects of Cisplatin (Table 2 and Figure 2).

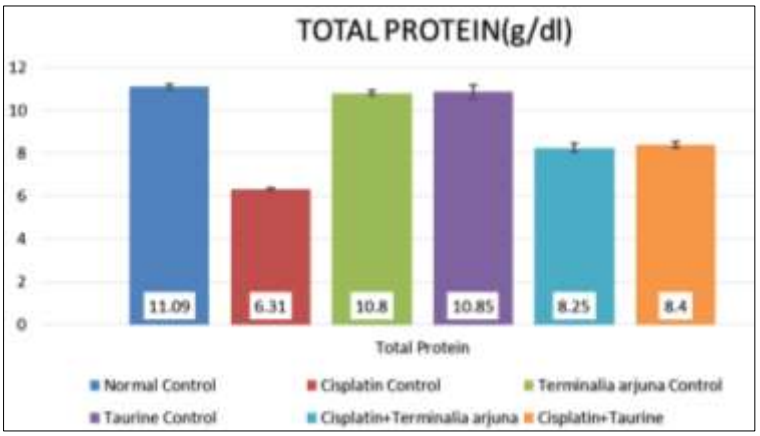

Fig 2: Total protein concentration (g/dl) in different groups of rats

Prothrombin time

In Group 2, the prothrombin time (sec) is significantly ($P < 0.05$) increased compared to group 1, 3 and 4. Administration of aqueous leaf extract of *Terminalia arjuna* (group 5) and taurine (group 6) significantly ($P < 0.05$) decreased values when compared to group 2 (Table 2 and Figure 3).

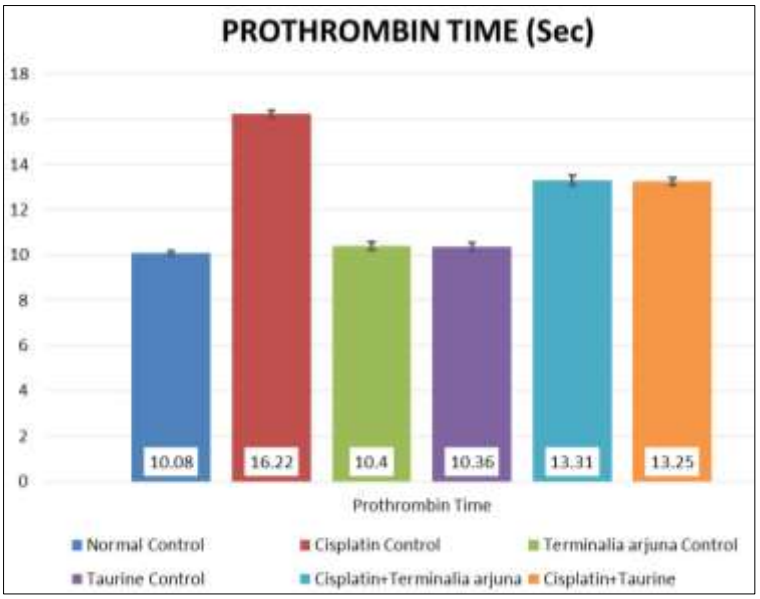

Fig 3: Prothrombin time (sec) in different groups of rats

Table 2: Average body weights (g), total protein (g/dl) and prothrombin time (sec) of different groups of rats

Group	Day 7 Body weight	Day 14 Body weight	Total protein	Prothrombin time
1.Normal Control	222.96 ± 0.51 ^{a}	231.31 ± 0.38 ^{a}	11.09 ± 0.11 ^{a}	10.08 ± 0.11 ^{c}
2.Cisplatin Control	164.40 ± 1.23 ^{d}	153.85 ± 0.96 ^{c}	6.31 ± 0.07 ^{c}	16.22 ± 0.15 ^{a}
3.*Terminalia arjuna*	220.15 ± 0.49 ^{b}	229.81 ± 0.56 ^{a}	10.80 ± 0.12 ^{a}	10.40 ± 0.17 ^{c}
4.Taurine	221.56 ± 0.53 ^{ab}	230.38 ± 0.52 ^{a}	10.85 ± 0.32 ^{a}	10.36 ± 0.18 ^{c}
5.Cisplatin + *Terminalia arjuna*	204.91 ± 0.82 ^{c}	213.81 ± 0.49 ^{b}	8.25 ± 0.19 ^{b}	13.31 ± 0.22 ^{b}
6.Cisplatin + Taurine	206.48 ± 0.73 ^{c}	214.63 ± 0.33 ^{b}	8.40 ± 0.14 ^{b}	13.25 ± 0.16 ^{b}

Table 4.1: Average body weights (g) of different groups of rats Values are Mean + SE (n =6); One way ANOVA with Duncan's post hoc test (SPSS). Means with different alphabets as superscripts differ significantly ($P < 0.05$) among the groups (Vertically).

Discussion

Cisplatin (5 mg/kg, i.p) administration to the rats resulted in noticeable nephrotoxicity and hepatotoxicity in Cisplatin control rats, which was manifested by frequent urination, diarrhoea, abnormal posture and ataxia as compared to normal control rats. Dissection of these animals indicated that the loss of the body weights were due to loss of skeletal muscles and adipose tissue as previously suggested by [22]. This weight loss observed in the Cisplatin treated group may be because of reduced appetite and enhanced catabolic rate, which are considered as the obvious side effects of chemotherapy. It could also be due to Cisplatin induced dysfunction of the gastrointestinal system. The treatment affects the target organs like kidney and liver as evidenced by their elevated

functional markers in the serum of the Cisplatin treated group [23]. In, contrast, There was no difference between the body weights before and after the experiments among the groups [24, 25, 26]. Decrease in the body weights was also observed due to gastrointestinal toxicity [27, 28].

In the present study, the body weights of rats in toxic control group 2 were significantly reduced when compared to non-toxic control groups (1, 3 and 4) at respective time intervals. The decrease in body weights in Cisplatin group is attributed to oxidative stress and excess ROS generation, which leads to altered metabolism [29]. The decrease in body weight of the animals in this study correlate with the reduced food intake observed during the experimental period. Weight gain observed in the treated animals (groups 5 and 6) may be due to restoration of antioxidant defenses.

The present study also revealed a significant alteration in the total protein in toxic control group 2 when compared to the non-toxic control groups (1, 3 and 4), which might be attributed to the impairment of liver function due to Cisplatin administration resulting in oxidative stress and decreased capacity of liver to synthesize proteins. Cisplatin administration caused a significant decrease in serum proteins by its impaired synthesis in liver [30]. Total protein levels reduced in hepatotoxic/nephrotoxic conditions resulted from the faulty protein biosynthesis [31, 32]. Cisplatin administration produced a significant decrease in total proteins that reflect its interaction with cell membrane, leading to altered cell membrane permeability and alteration of functional integrity in the kidney [33]. These significant decrease in total protein due to the direct effect of the Cisplatin on protein metabolism or due to enhanced proteolytic activity as a consequence of increased metabolic demands following exposure to Cisplatin [34]. There was significant increase in the total protein of treatment groups (5 and 6) compared to the values of toxic control group.

Tissue injury induced by Cisplatin injection may explain the marked changes in prothrombin time (Pt). In addition, a significant gradual increase in the Pt started 1 day after Cisplatin injection and peaked after 4 days. The increase in Pt was accompanied with significant decrease in the platelet count. This could be attributed to activation of platelets as a result of activation of coagulation cascade activated platelets liberate chemotactic substances and adhesion molecules, which attract coagulation factors, inflammatory mediators, leukocytes and other platelets from blood to the injured tissue causing thrombocytopenia [35]. The hematological parameters are fit with the biochemical and histological results which confirmed by the renal impairment 4 days after Cisplatin injection [36].

Conclusion

In conclusion, Cisplatin caused the formation of free radicals in the liver and kidney by reducing the antioxidant indices. However, aqueous leaf extract of *Terminalia Arjuna* and taurine supplementation to Cisplatin injected rats exhibited no adverse effects indicating its protective antioxidant property. Thus, present investigation confirmed the protective role of *Terminalia Arjuna* and taurine against Cisplatin toxicity.

Acknowledgment

The authors are thankful to College of veterinary science, PVNR TVU, Rajendranagar, Hyderabad for providing necessary facilities for research work.

References

1. Crona DJ, Faso A, Nishijima TF, McGraw KA, Galsky MD, Milowsky MI. A systematic review of strategies to prevent cisplatin-induced nephrotoxicity. The oncologist 2017;22(5):609.
2. Yadav YC. Effect of cisplatin on pancreas and testies in Wistar rats: biochemical parameters and histology. Heliyon 2019;5(8):e02247.
3. Xu Z, Brodbelt JS. Differentiation and distributions of DNA/cisplatin crosslinks by liquid chromatography-electrospray ionization-infrared multiphoton dissociation mass spectrometry. Journal of the American Society for Mass Spectrometry 2013;25(1):71-79.
4. Tsang RY, Al Fayea T, Au HJ. Cisplatin overdose. Drug safety 2009;32(12):1109-1122.
5. Al Majed AA. Carnitine deficiency provokes cisplatin-induced hepatotoxicity in rats. Basic & clinical pharmacology & toxicology 2007;100(3):145-150.
6. Rehman M U, Ali N, Rashid S, Jain T, Nafees S, Tahir M *et al*. Alleviation of hepatic injury by chrysin in cisplatin administered rats: probable role of oxidative and inflammatory markers. Pharmacological Reports 2014;66(6):1050-1059.
7. Ibrahim A, Abd Eldaim MA, Abdel Daim MM. Nephroprotective effect of bee honey and royal jelly against subchronic cisplatin toxicity in rats. Cytotechnology 2016;68(4):1039-1048.
8. Valentovic MA, Ball JG, Brown JM, Terneus MV, McQuade E, Van Meter S *et al*. Resveratrol attenuates cisplatin renal cortical cytotoxicity by modifying oxidative stress. Toxicology in Vitro 2014;28(2):248-257.
9. Ramya B, Anjaneyulu Y, Reddy AG, Madhuri D, Lakshman M, Shivakumar P. Protective role of turmeric on histological, ultrastructural and sero-biochemical changes in cisplatin-induced nephrotoxicity in female rats. Veterinary World 2013;6(11):865.
10. Nematbakhsh M, Ashrafi F, Pezeshki Z, Fatahi Z, Kianpoor F, Sanei MH *et al*. A histopathological study of nephrotoxicity, hepatoxicity or testicular toxicity: Which one is the first observation as side effect of Cisplatin-induced toxicity in animal model. Journal of nephropathology 2012;1(3):190.
11. Pezeshki Z, Khosravi A, Nekuei M, Khoshnood S, Zandi E, Eslamian M *et al*. Time course of cisplatin-induced nephrotoxicity and hepatotoxicity. Journal of nephropathology 2017;6(3):163.
12. Un H, Ugan RA, Kose D, Bayir Y, Cadirci E, Selli J. A novel effect of Aprepitant: Protection for cisplatin-induced nephrotoxicity and hepatotoxicity. European Journal of Pharmacology 2020, 173168.
13. Sudhavani V, Chinnikrishnaiah V, Raghu Moorthy V, Raghavendra HG, Ranganayakulu D. Merremiaemarginata burmprotects against cisplatin induced nephrotoxicity rats. Journal of Advances in Drug Research 2010;1:2230-7761.
14. Dwivedi S. Terminalia arjuna Wight & Arn. a useful drug for cardiovascular disorders. Journal of ethnopharmacology 2007;114(2):114-129.
15. Ahmad MS, Ahmad S, Gautam B, Arshad M, Afzal M. Terminalia arjuna, a herbal remedy against environmental carcinogenicity: an in vitro and in vivo study. Egyptian Journal of Medical Human Genetics 2014;15(1):61-68.
16. Bhattacharjee B, Pal PK, Ghosh AK, Mishra S,

Chattopadhyay A, Bandyopadhyay D. Aqueous bark extract of Terminalia Arjuna protects against cadmium-induced hepatic and cardiac injuries in male Wistar rats through antioxidative mechanisms. Food and Chemical Toxicology 2019;124:249-264.
17. Das J, Sil PC. Taurine ameliorates alloxan-induced diabetic renal injury, oxidative stress-related signaling pathways and apoptosis in rats. Amino acids 2012;43(4):1509-1523.
18. Manna P, Sinha M, Sil PC. Taurine plays a beneficial role against cadmium-induced oxidative renal dysfunction. Amino Acids 2009;36(3):417-428.
19. Ghosh J, Das J, Manna P, Sil PC. Taurine prevents arsenic-induced cardiac oxidative stress and apoptotic damage: Role of NF-κB, p38 and JNK MAPK pathway. Toxicology and applied pharmacology 2009;240(1):73-87.
20. Roy A, Manna P, Sil PC. Prophylactic role of taurine on arsenic mediated oxidative renal dysfunction via MAPKs/NF-κ B and mitochondria dependent pathways. Free radical research 2009;43(10):995-1007.
21. Raj S, Singh H, Trivedi R, Soni V. Biogenic synthesis of AgNPs employing Terminalia arjuna leaf extract and its efficacy towards catalytic degradation of organic dyes. Scientific Reports 2020;10(1):1-10.
22. Basuony M, Tousson E, Massoud A, Saad S. Preventive/Ameliorative Effects of Ginseng Aqueous Extract on Cisplatin-induced Hepatic and Renal Injuries. Asian Oncology Research Journal 2020, 31-39.
23. Hassan I, Chibber S, Naseem I. Ameliorative effect of riboflavin on the cisplatin induced nephrotoxicity and hepatotoxicity under photoillumination. Food and Chemical Toxicology 2010;48(8-9):2052-2058.
24. de Oliveira Mora L, Antunes LMG, Francescato HDC, Bianchi MDLP. The effects of oral glutamine on cisplatin-induced nephrotoxicity in rats. Pharmacological Research 2003;47(6):517-522.
25. Bentli R, Parlakpinar H, Polat A, Samdanci E, Sarihan ME, Sagir M. Molsidomine prevents cisplatin-induced hepatotoxicity. Archives of medical research 2013;44(7):521-528.
26. Cagin YF, Erdogan MA, Sahin N, Parlakpinar H, Atayan Y, Polat A *et al*. Protective effects of apocynin on cisplatin-induced hepatotoxicity in rats. Archives of medical research 2015;46(7):517-526.
27. Devi PS, Shyamala DC. Protective effect of quercetin in cisplatin-induced cell injury in the rat kidney. Indian journal of pharmacology 1999;31(6):422.
28. Maheshwari RA, Sailor GU, Patel L, Balaraman R. Amelioration of cisplatin-induced nephrotoxicity by statins. Indian journal of pharmacology 2013;45(4):354.
29. Kumar M, Dahiya V, Kasala ER, Bodduluru LN, Lahkar M. The renoprotective activity of hesperetin in cisplatin induced nephrotoxicity in rats: molecular and biochemical evidence. Biomedicine & Pharmacotherapy 2017;89:1207-1215.
30. Diab AA, Zahra MH, Hendawy AA, Hamza RZ, Mekky GA. Hepatoprotective effect of curcumin and vitamin C against cisplatin induced oxidative stress and toxicity in albino rats. Journal of American Science 2014;10(11):37-48.
31. Renugadevi J, Prabu SM. Naringenin protects against cadmium-induced oxidative renal dysfunction in rats. Toxicology 2009;256(1-2):128-134.
32. Parameshappa B, Ali Basha MS, Sen S, Chakraborty R, Kumar GV, Sagar GV *et al*. Acetaminophen-induced nephrotoxicity in rats: protective role of Cardiospermum halicacabum. Pharmaceutical biology 2012;50(2):247-253.
33. Sen S, De B, Devanna N, Chakraborty R. Cisplatin-induced nephrotoxicity in mice: protective role of Leea asiatica leaves. Renal failure 2013;35(10):1412-1417.
34. Al Malki AL, Sayed AAR. Thymoquinone attenuates cisplatin-induced hepatotoxicity via nuclear factor kappa-β. BMC complementary and alternative medicine 2014;14(1):1-8.
35. Gresele P, Momi S, Malvestiti M, Sebastiano M. Platelet-targeted pharmacologic treatments as anti-cancer therapy. Cancer and Metastasis Reviews 2017;36(2):331-355.
36. Ewees MG, Messiha BA, AboSaif AA, Bayoumi A, Abdel Bakky MS. Interference with coagulation cascade as a novel approach to counteract cisplatin-induced acute tubular necrosis; an experimental study in rats. Frontiers in pharmacology 2018;9:1155.