Bulletin of Environment, Pharmacology and Life Sciences
Bull. Env. Pharmacol. Life Sci., Vol 10 [12]November 2021 : 80-84

Online ISSN 2277-1808
Journal's URL:http://www.bepls.com
CODEN: BEPLAD

ORIGINAL ARTICLE **OPEN ACCESS**

Nephroprotective and antioxidant activity of *Albizzia lebbeck.*

Shirode D.S.[1]*, Chaudhari P. M.[1], Bindurani L.G.P. Ram[2] , Agarwal A[3],
[1]Dept. of Pharmacology, Dr. D. Y. Patil College of Pharmacy, Akurdi, Pune India
[2]Faculty of Pharmacy, Vishwakarma University, Kondhwa (BK), Pune-411048 Maharashtra
[3]Faculty of Pharmacy, Mansarover Global University, Billkisganj, Sehore, Madhya Pradesh - 466001
*For correspondence to: Dr. D. S. Shirode
E-mail address: devendrashirode@dyppharmaakurdi.ac.in

ABSTRACT
Nephrotoxicity is the renal injury due to medication. More than 50 % of complicated patients develop acute kidney injury after frequent use of drugs. Various drugs as adverse effect are responsible for nephrotoxicity like aminoglycoside antibiotics, anticancer drugs etc. The present study was designed to evaluate the nitric oxide scavenging activity and nephroprotective activity of 70% ethanolic extract of leaves of Albizzia lebbeck (EELAL) against Cisplatin (6mg/kg.i.v.) induced nephrotoxicity in albino rats. Preliminary phytochemical screening, in vitro antioxidant model, acute oral toxicity was performed. The degree of nephrotoxicity/protection was determined by measuring the level of Physical parameter (wet kidney weight), biochemical markers (BUN and serum creatinine) and in vivo antioxidant parameters i.e. Glutathione (GSH) and Lipid peroxidation (LPO). The 70 % EELAL significantly decrease the activity of tissue lipid peroxidation and Biochemical markers (BUN and serum creatinine) while it significantly increased in nitric oxide scavenging activity and tissue GSH levels in dose dependent manner. Histopathology studies and physical parameter also supported these results. The nephroprotective and antioxidant properties may be attributed to the polyphenolic compounds like flavonoids and tannins that are present in the leaves of Albizzia lebbeck
***Keywords**: Albizzia lebbeck, Cisplatin, Antioxidant, Nephrotoxicity*

Received 28.08.2021 Revised 18.10.2021 Accepted 12.11.2021

INTRODUCTION

There are various drugs such as Cisplatin, Gentamicin and Paracetamol, which are used for their therapeutic effects but they produce nephrotoxicity as adverse effect. Gentamicin causes nephrotoxicity about 13-30% to treated patients [1]. Overdose of paracetamol lead to kidney failure in experimental animals and humans in severe case to death [2]. Cisplatin act as chemotherapeutic agent for solid tumors but it also causes the serious adverse effect such as nephrotoxicity [3]

As natural products show significant contribution and less adverse effect for the treatment of various diseases. On the basis of literature survey and availability of plant to native practitioner, *Albizzia lebbeck* plants were selected for nephroprotective activity. *Albizzia lebbeck* Benth belongs to family Mimosaceae. It useful in opthalmia, aphrodisiac [4], allergic disorders [5], chronic cough, bronchitis [6], inflammation, scrofula, skin disease, leprosy, leucoderma, seminal weakness, ophthalmopathy and poisoning. The leaves of the plant *Albizzia lebbeck* are rich in vicenin II, β-sitosterol, echinocystic acid and flavon. etc [5]. The modern literature revealed that the plant is reported to possess nootropic [7, 8], anxiolytic [8], anticonvulsant [9, 10], antifertility [11], antidiarrheal activity [12], anti-inflammatory (bark) [13, 14], antiulcer [15] and hepatoprotective [16].

The purpose of the present study was to evaluate the ability of 70% ethanolic extract of leaves of *Albizzia lebbeck* (EELAL) to protect renal tissue against Cisplatin induced nephrotoxicity in rats.

MATERIAL AND METHODS

Plant Material

The *Albizzia lebbeck* leaves were collected from fields of Anand, Gujarat. It was authenticated by Prof. G.C. Jadeja, Dept of Agricultural Botany, Anand Agricultural University. The 70% ethanolic extract was prepared by using 70% ethanol in a soxhlet apparatus after de-fatting with petroleum ether. Preliminary phytochemical result of 70% EELAL exhibited the presence of saponins glycoside, tannin and flavonoids in it. So EELAL was further selected for acute oral toxicity and nephroprotective study.

Animals

Female mice (18-25 g) and either sex of Wistar albino rats (150-220g) were used for the study, Approval from the institutional animal Ethical committee (Reg. no. 1554/PO/a/11/CPCSEA) was obtained for preclinical studies as per the CPCSEA norms.

Acute Toxicity studies

OECD Guide line no 420 was used for the acute oral toxicity studies. Groups of 6 mice were administered test drug by oral route in the range of 2000-300 mg/kg and mortality was observed after 24 hr.

***In vitro* Antioxidant activity**

Sodium nitroprusside (1ml of 10mM) were mixed with 1ml of EELAL at different concentration (20-100 µg/ml) in phosphate buffer (pH 7.4). The mixture was incubated at 25°C for 150 min. Griess's reagent (1 ml) was added in 1 ml of the incubated solution. Absorbance was read at 546 nm [17]

Cisplatin induced nephrotoxicity

In this model, four groups of rat animals were prepared and each group contained six rat animals. Saline (1ml/kg, p.o) was administrated to group I and II for 7 days. The group III and IV were treated with EELAL 100 mg/kg p.o and 200mg/kg p.o for 7 days, respectively. On 2nd day, 30 min after saline, 100 mg/kg EELAL and 200 mg/kg EELAL administration to group II, III and IV respectively, received cisplatin (6mg/kg, i.v). Saline, 100 mg/kg EELAL and 200 mg/kg EELAL in groups II, III and IV respectively was administered once a day on days 1, 4, 5 & 6 and twice a day on days 2 and 3 [18]. The blood samples were collected from retro-orbital plexus and subjected for estimation of biochemical markers. The animals were sacrificed and kidney samples were collected for estimation of GSH, lipid peroxidation, physical parameter and histopathological studies.

Parameter assess for the renal functions

Wet kidney weight**,** Body weight, Blood urea [19] and Serum creatinine [20], GSH, lipid peroxidation and histopathological studies

GSH estimation

GSH was estimated as per Ellamn procedure [21]. 1 gm tissue samples were homogenized in 10 ml, 10% TCA. After centrifugation at 3000 rpm for 10 minutes, 0.5 ml supernatant was added to 2 ml disodium hydrogen phosphate solution (0.3 M). The absorbance was determined at 412 nm after mixing solution with 0.2 ml dithiobisnitrobenzoate (0.4 mg/ml in 1% sodium citrate).

In vivo lipid peroxidation

Lipid peroxidation was measured by monitoring thiobarbituric reactive substance formation [22]. Stock solution of TCA (15% w/v) –TBA (0.375% w/v) - HCl (0.25 N) reagent was prepared. Added 1.0 ml biological sample (0.1-2.0 mg of membrane protein or 0.1-0.2 µmol of lipid phosphate) with 2.0 ml TCA-TBA- HCl and mix thoroughly. The boiling water bath was used to heat the solution for 15 minutes.

After centrifugation for 10 minutes at 100 rpm, precipitate was removed. The absorbance was determined at 535 nm.

Statistical analysis

Results were expressed as mean ± SEM, (n=6). One way analysis of variance (ANOVA) was performed using Tukey-Kramer Multiple Comparisons method for Statistical analysis. P value less than 0.05 was considered to be statistically significant. *P<0.05, **<0.01 and ***<0.001, when compared with toxicant group.

RESULTS AND DISCUSSION

Acute Toxicity studies

During the acute toxicity study, all the animals died at 2000 mg/kg and one animal died at 300 mg/kg. So, as per the OECD guideline 420, 1/10th and 1/5th dose (100 mg/kg and 200 mg/kg) were selected using LD_{50} cut off dose for further studies.

Antioxidant activity

The in vitro antioxidant studies of the EELAL demonstrated dose dependant nitric oxide scavenging activity (Table 1).

Effect of EELAL on Cisplatin induced nephrotoxicity

Cisplatin administration exhibited a marked decrease in body weight (-18.3%), tissue GSH level (0.416±0.018) and increased kidney weight (1.09±0.065) and lipid peroxidation (0.439±0.013) which was supported by a significant increase in serum markers like blood urea (75.45±1.89) and serum creatinine (2.11±0.062). Administration of EELAL normalized the raised kidney weight, blood urea, serum creatinine, tissue lipid peroxidation and prevented depleted tissue GSH level and body weight (Table 2).

Cisplatin administration caused glomerular congestion, severe interstitial with inflammatory cells, suggesting initiation of necrosis development. These histopathological assessments were dose dependently reversed with the treatment of EELAL. Administration of EELAL normalized the physical parameter (kidney weight), Biochemical parameters (blood urea, serum creatinine), in vivo antioxidant parameters (tissue LPO and tissue GSH) and body weight. Hence, the EELAL showed nephroprotective effect in cisplatin induced nephrotoxicity in rat model.

Table 1: Nitric oxide radical scavenging activity of EELAL.

Groups	Absorbance Mean ± SEM	% Inhibition
Control	0.305 ± 0.002	--
Control + standard 25 µg	0.108± 0.0025***	64.59
Control + EELAL 20 µg	0.292 ± 0.003***	4.26
Control + EELAL 40 µg	0.261 ± 0.001***	14.43
Control + EELAL 60 µg	0.229 ± 0.003***	24.92
Control + EELAL 80 µg	0.196 ± 0.001***	35.78
Control + EELAL 100 µg	0.165 ± 0.002***	45.90

Values are the mean ± S.E.M., n=3; Significance *** P<0.001, compared to control.
Std: Sodium metabisulphate

Table 2: Effect of 70% EELAL on GSH, lipid peroxidation, physical and biochemical parameters in Cisplatin induced renal damage in rats

Gr. (n=6)	Treatment regimen	Kidney weight (g/100g)	Change in b.w. (%)	Blood urea (mg/dl)	Serum creatinine (mg/dl)	Glutathione (Mean ± SEM)	Lipid peroxidation (Mean ± SEM)
I	Vehicle treatment (Negative control)	0.6835 ± 0.02038	8.3 ± 1.667	27.84 ± 0.7040	0.63 ± 0.032	0.812 ± 0.016	0.176 ± 0.0078
II	Cisplatin 6 mg/kg i.v. on 2nd day (Positive control)	1.09 ± 0.06591	-18.3 ± 1.667	75.45 ± 1.898	2.11 ± 0.062	0.416 ± 0.018	0.439 ± 0.0139
III	Cisplatin 6 mg/kg i.v. on 2nd day +EELAL 100 mg/kg p.o.	0.824 ± 0.06***	-10.83 ± 1.537*	45.61 ± 2.54***	1.45 ± 0.05***	0.528 ± 0.028* (26.9%)	0.310 ± 0.01*** (29.3%)
IV	Cisplatin 6 mg/kg i.v. on 2nd day + EELAL 200 mg/kg p.o. s	0.712 ± 0.02***	-6.6 ± 1.05***	38.12 ± 1.36***	0.91 ± 0.02***	0.659 ± 0.027** (58.4%)	0.245 ± 0.01*** (44.1%)

Values are the Mean ± S.E.M. of six rats / treatment, Significance *P<0.05, **P <0.01 and *** P<0.001,
b.w. – Body weight

DISCUSSION

Anticancer drug, cisplatin generates free radicals and increase lipid peroxidation lead to nephrotoxicity. Even report also suggests Cisplatin cause cytotoxicity effects due to inhibition of antioxidant activities and generation of reactive oxygen species [23]. Cisplatin increases the nitric oxide production through arginine metabolism [24]. In the present study, EELAL administration significantly decreased the lipid peroxidation and increased the tissue GSH level and NO radical scavenging activities *(in-vitro* studies). Therefore the nephroprotective activity of the EELAL may be recognized to the antioxidant activity of the plant.

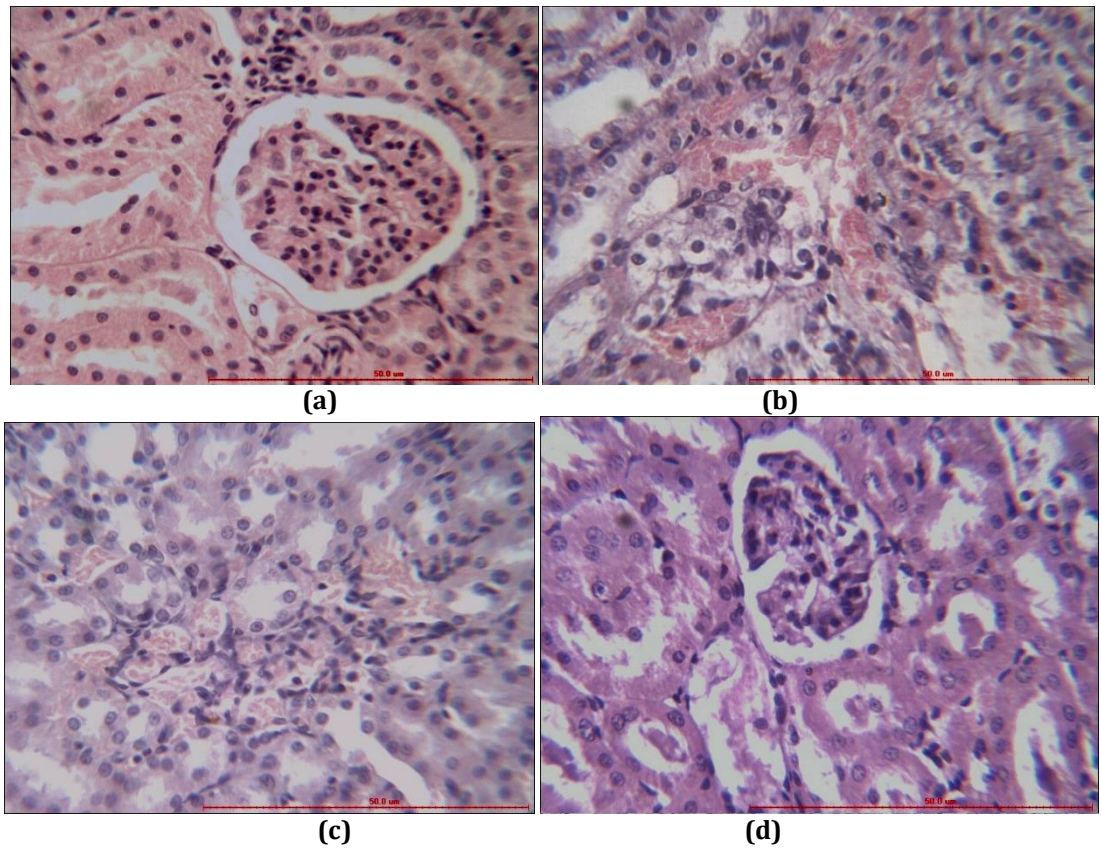

Fig. 1. Photomicrograph of kidney architecture of **(a)** Normal Control **(b)** Cisplatin intoxicated showing severe interstitial congestion. **(c)** Cisplatin intoxicated + 100 mg/kg of EELAL, showing Recovery of Interstitial congestion. **(d)** Cisplatin intoxicated + 200 mg/kg of EELAL, showing interstitial congestion like normal (H x E 200)

CONCLUSION

EELAL exhibited the nitric oxide scavenging and nephroprotective activity. It may be due to the presence of the polyphenol compounds of plant, namely tannins and flavonoids. However, further, investigation on Isolation, characterization and formulation of active compound is required to validate the antioxidant principles and nephroprotective property.

ACKNOWLEDGEMENTS

We are thankful to Dr. D. Y. Patil College of Pharmacy, Akurdi, Pune for their constant support and providing all the facilities to carry out this research work.

REFERENCES

1. Lorz, C., Justo, P., Sanz, A., Sufria, D., Egido. J., Ortiz, A. (2004). Paracetamol Induced Renal Tubular Injury: A Role for ER Stress. J. Am. Soc. Nephrol., 15: 380-389.
2. Ali, B.H., Ismail, T.H.B., Bashir, A.A. (2001). Sex difference in the susceptibility of rats to gentamicin nephrotoxicity: Influence of gonadectomy and hormonal replacement therapy. Indian J. Pharmacol., 33: 369-373.
3. Kunihiko, S., Kazuto, M., Kazutaka, M., Yoshinori, I., Kazuhiko, T., Hideki, H., Ryozo, O. (2002). Protection by a radical scavenger edaravone against cisplatin-induced nephrotoxicity in rats. Eur. J. Pharmacol., 451(2): 203-208.
4. Kritikar, K.R., Basu, B.D. (1998). Indian Medicinal Pants, Lalit Mohan Babu, Allahabad. 936.
5. Asolkar, L.V., Kakkar, K.K. (1965). Second supplement to Glossary of Indian Medicinal Plants with active principles. 36.
6. Vaidyaratnum, P.S. (1995). Indian Medicinal Plants, Arya Vaidyasala, Kottakkal, Orient Longman. 81.
7. Chintawar, S.D., Somani, R.S., Kasture, V.S., Kasture, S.B. (2002). Nootropic activity of *Albizzia Lebbeck* in mice. J. Ethnopharmocol, 81: 299
8. Une, H.D., Sarveiya, V.P., Pal, S.C., Kasture, V.S., Kasture, S.B. (2001). Nootropic & anxiolytic activity of Saponins of *Albizzia lebbeck* leaves. Pharmacology Biochemistry and Behavior, 69: 439-444.
9. Kasture, V.S., Chopade, C.T., Deshmukh, V.K. (2000). Anticonvulsant activity of *Albizzia lebbeck, Hibiscus rosa sinesis* and *Butea monosperma* in experimental animals. Journal of Ethanopharmacology, 71: 65-75.

10. Kasture, V.S, Kasture, S.B., Pal, S.C. (1996). Anticonvulsant activity of *Albizzia lebbeck* leaves. Indian J. Exp. Biol., 34: 78-80
11. Gupta, R.S., Kachhawa, J.B., Chaudhary, R. (2004). Antifertility effects of methanol pod extract of *Albizzia lebbeck* (L) Benth in male rats. Asian J. Androl., 6: 155-159.
12. Besra, S.E, Gomes, A., Chaudhury, L., Vedasiromoni, J.R., Ganguly, D.K. (2002) Antidiarrhoeal activity of seed extract of *Albizzia lebbeck* Benth. Phytotherapy Res., 16: 529-533.
13. Pramanik, K.C., Bhattacharya, P., Chatterjee, T.K., Mandal, S.C. (2005). Anti-inflammatory activity of methanol extract of *Albizzia lebbeck* bark. European Bulletin of drug research, 13: 71-75.
14. Shirode, D.S., Roy, S.P., Patel, T., Rajendra, S.V., Jyothi, T.M., Setty, S.R. (2008). Antiinflammatory activity of ethanolic extract of *Albizzia lebbeck* leaves and *Madhuca longifolia* bark. Int. J. Pharmacol. Biol. Sci., 2 (3): 127-130.
15. Shirode, D.S., Patel, T., Roy, S.P., Rajendra, S.V., Jyothi, T.M., Setty, S.R. (2008). Antiulcer properties of 70 % ethanolic extract of leaves of *Albizzia lebbeck.* Phcog. mag., 4 (15): 228.
16. Shirode, D.S., Roy, S.P., Patel, T., Rajendra, S.V., Jyothi, T.M., Setty, S.R. (2008). Evaluation of hepatoprotective effect of leaves of seventy percent ethanolic extract of *Albizzia lebbeck* in paracetamol induced experimental hepatic damage. Plant Archives, 8 (2): 797-801.
17. Mondal, S.K., Chatraborty, G., Gupta, M., Mazumder, U.K. (2004). Invitro antioxidant activity of *Dispyros malabarica* Kostal bark. Indian J. Exp. Biol., 44: 39-44.
18. Corcostegul, R., Labeaga, L., Arteche, J.K., Orjales, A., (1998). Protective effect of hidrosmin against cisplatin induced acute nephrotoxicity in rats. J. Pharm. Pharmacol., 4: 465-467.
19. Tiftany. T.O., Jansen, J., Burtis, C.A., Overton J.B., Scott, C.D. (1972). Clin. Chem., 18: 829.
20. Henry, J.B., Young, D.S., Tietz, N.W., Vasildes, J. (1972). Can, Chem. 18
21. Aykae, G., Vysal, M., Yalein, A.S., Kocak, T.N., Sivas, A., Oz, H. (1985). The effect of chronic ethanol ingestion on hepatic lipid peroxide, Glutathione, glutathione peroxidase and glutathione transferase in rats. Toxicology, 36: 71-76.
22. Buege, A. J., Steven, A.D. (1978). Microsomal lipid peroxidation. London: Moury Kleiman Co., 302.
23. Devi, P., S., Shyamala, D.C.S. (1999). Protective effect of quercetin in cisplatin induced cell injury in the rat kidney. Ind. J. Pharmacol., 31: 422-426.
24. Annie, S., Rajagopal, P.L., Malini, S. (2005). Effect of *Cassia auriculata* Linn. Root extract on cisplatin and gentamicin-induced renal injury. Phytomedicine, 12 (8): 555-560.

CITATION OF THIS ARTICLE

Shirode D.S., Chaudhari P. M., Bindurani L.G.P. Ram , Agarwal A. Nephroprotective and antioxidant activity of *Albizzia lebbeck..* Bull. Env. Pharmacol. Life Sci., Vol 10[12] November 2021 : 80-84.