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Research Article

Studies on Nephroprotective Effect of Methanolic and Aqueous Extract of Bark of *Pithecellobium Dulce* Roxb. in Rats

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ABSTRACT

Kidney failure is a major problem of worldwide proportions. Kidney injury is a sudden loss of kidney function resulting in the accretion of waste materials such as creatinine and urea in the body. There are various agents that exert nephrotoxic effects through distinctive morbific defence mechanisms. Gentamicin, cisplatin and paracetamol are among common nephrotoxicity agents. In recent years, natural compounds are being expanded used in the treatment of kidney diseases. With this backdrop the study has been designed with the aim to studies on nephroprotective effect of methanolic and aqueous extract of bark of *Pithecellobium Dulce* Roxb. in rats. The methanolic and aqueous extracts of bark of *P. Dulce* was studied for nephroprotective effect in female Wistar rats against cisplatin, gentamicin and paracetamol induced nephrotoxicity, by estimating serum creatinine, blood urea and serum albumin levels. The dried bark of *P. Dulce,* Roxb (*Leguminosae*), contains tannins, flavonoids, triterpenoids, beta-sitosterol and saponin glycoside. Treatments with the methanolic and aqueous extracts could significantly ($P < 0.001$) reduce the elevated serum levels of creatinine, blood urea and serum albumin. *P. Dulce* bark extracts significantly prevented the physical, biochemical, histological and structural changes induced by cisplatin, gentamicin and paracetamol in the kidney. The present study reports with the exploration of *P. Dulce* bark extracts possessed *implicit* nephroprotective activity. This activity may be attributed due to the abundant phytoconstituents of the plant extracts.

Introduction

Nephrosis is a chronic disorder of environmental toxicant, noxious chemicals, and drugs, including antibiotics, drugs used in chemotherapy like cisplatin, almighty alter the structure or internal functioning of several organs and create harmful effects on kidney, heart, pancreas and intestine etc.[1] Polygenic disorder like diabetes is the most common cause of kidney failure, accounting for nearly 44 percent of the diabetic cases.[2]

Even when diabetes is controlled, the disease can lead to kidney failure. Many people with diabetes do not develop chronic kidney disease (CKD) that is severe enough to progress to kidney failure. Nearly 24 million people in the United States have diabetes.[3] Nephrogenic fibrosing dermopathy is a common significance of chronic kidney failure. In all cases, the expanse of production and deposition of extracellular matrix (ECM) proteins in the kidney firmly correlates with disease asperity, impairment of renal function and subsequent progression to end-stage renal disease (ESRD).[4]

Kidney being one of the indispensable organs of human body performing the function of removing toxic substances needs protection for healthy life. As stated above, the prevalence of the kidney disorders needs to be seriously viewed. Ethno-medicinal plants can be acclimated to advice apprehend the charge for dialysis by treating the causes and after effect of kidney failure, as well as reducing the many adverse effect of dialysis. In the established pharmacopoeia, *P. dulce* bark reduced into powder percolated in alcohol or water is used to pyrexia and diarrhea, injury chancres, sickness, and seizure.[5] *P. dulce* prohibited Naja kaouthia venom activities by

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precipitating venom proteins due to condensed tannins.[6] In fact, relating to aqueous and alcoholic stem bark extract contains excessive amounts of phenolic compounds including, flavonoids and pro-anthocyanidin. The dried bark of *P. dulce*, (*Leguminosae*), is tree indigenous to the America, central Asia and India. The bark yields 37% tannins of the catechol type while the leaves yield quercetin, kaempferol, dulcitol and afezilin.[7] The bark of *P. dulce* is utilized for treatment of liver disorders.[8] It also acts probably by scavenging the free radicals and inhibition of generation of oxygen species.[9] The drug has not yet explored for the treatment of kidney disorders and there is a high probability that the drug would prove to be beneficial in offering protection against the kidney failure in patients suffering from diabetes, hypertension and cardiac disorders.

The bark has found to lower the blood glucose levels of alloxan induced diabetes in rats.[10] With this background the study has been designed with the aim to evaluate the bark of *P. Dulce,* Roxb (*Leguminosae*), for its nephroprotective activity so that the drug can be utilized to protect kidneys, as the organ needs serious attention in the contemporary lifestyle.

Materials and Methods

Plant Material

The barks of *P. Dulce* (family- *Leguminosae*) were collected from Chopda area of the district-Jalgaon, during the months of April to June and authenticated by department of Biodiversity and Palaeobiology, Maharashtra Association for the Cultivation of Science, Agharkar Research Institute, Pune–411004 (Certificate number AUTH 19-129 and Reference No – COPS/2661/2018-19). The sample has been critically studied with the help of microscope, organoleptic characters and TLC profile.

Drugs and Chemicals

The following chemicals were used for the study, cisplatin injection (Cismax 50 GLS Pharma limited), gentamicin injection (Gentalab, Laborate Pharmaceutical India Ltd), paracetamol (Paracip500, Cipla) urea estimation kit (Yucca Diagnostics), creatinine estimation kit (Delta Lab) and albumin kit (Pathozyme diagnostics).

Preparation of the Extract

The barks were shade dried for weeks. The dried barks were further chopped into small pieces and reduced to powder. *P. Dulce* bark powdered (250 gm) were defatted by extracting with petroleum ether (60–80°C), followed by extraction with methanol using soxhlet extractor. The methanolic extract was then concentrated rotary flash evaporator to a syrupy consistency. The residual solvent was removed by drying the extract in vacuum oven (yield - 25.5 gm). The aqueous extract of the plant was prepared by using the cold maceration process. The extract was then evaporated to dryness and the yield was noted (23.40 gm) and stored in air-tight, amber colored containers at room temperature for further proposed experimental studies.

Phytochemical Screening

The extract obtained was subjected to qualitative tests for identification of different phytoconstituents like alkaloids, tannins, glycosides, phenolic, flavonoids, proteins and steroids, by using standard and simple qualitative methods.[11]

Acute Toxicity Studies

Acute oral toxicity studies were performed as per OECD Guidelines 423. The study required female rats to be tested at the dose of 300 and 2000 mg/kg b.w.p.o. The animal was observed continuously first 30, 60, 120, 180 and 240 minutes after dosing, with special attention and once daily thereafter, for a total of 14 days. All animal showed no clinical signs of intoxication. Extracts did not cause morality in the female rats.

Pharmacological Study

Animals

Healthy female wistar rats (180–200 gm each) were used for the study. The animals were housed in polypropylene cages with stainless steel grill top and maintained under standard environmental conditions such as temperature ranging from 22 ± 3°C, relative humidity 55 ± 5% and illumination cycles set to 12 hours dark/light cycle. All the animals were fed with rodent pellet diet and water *ad libitum* under strict hygienic conditions. All procedures were performed in accordance with CPCSEA guidelines after approval from the Institutional Animal and Ethics Committee (IAEC) of the Crystal Biological solution [No. CRY/2021/006 & 2030/PO/RcBiBt/S/18/CPCSEA].

Evaluation of Nephroprotective Activity

Cisplatin-induced Nephrotoxicity[12]

Nephrotoxicity was induced by administering dose of cisplatin 5 mg/kg b.w.i.p. for seven consecutive days.

Gentamicin-induced Nephrotoxicity[13]

Nephrotoxicity was induced by administering dose of gentamicin 100 mg/kg b.w.i.p. for seven consecutive days.

Paracetamol-induced Nephrotoxicity[14]

Nephrotoxicity was induced by administering dose of paracetamol (2 g/kg) per orally on 7th day.

Treatment Schedule: The female Wistar rats (180-200 g) were divided into ten groups (n=6). The animals are given the following treatment in the study.

- *Group I:* Normal control (vehicle) group received gum acacia mucilage 2% w/v.
- *Group II:* Disease control group received single dose of cisplatin (5mg/kg b.w. *i.p.*) on day one.

- *Group III:* Cisplatin + methanolic extract of *P. Dulce* (500 mg/kg b.w.p.o) once a day, from day 1 to day 6.
- *Group IV:* Cisplatin + aqueous extract of *P. Dulce* (500 mg/kg b.w.p.o) once a day, from day 1 to day 6.
- *Group V:* Disease control group were injected with gentamicin (100 mg/kg b.w.i.p) for seven consecutive days.
- *Group VI:* Rats in this group were injected with gentamicin (100 mg/kg b.w.i.p) and administered methanolic extract of *P. Dulce* (500 mg/kg b.w.p.o) for seven consecutive days.
- *Group VII:* Rats in this group were injected with gentamicin (100 mg/kg b.w.i.p) and administered aqueous extract of *P. Dulce* (500 mg/kg b.w.p.o) for seven consecutive days.
- *Group VIII:* Disease control group were given paracetamol (2 g/kg b.w.p.o) per orally on 7th day.
- *Group XI:* Rats in this group the methanolic extract of *P. Dulce* (500 mg/kg b.w.p.o) were given for seven consecutive days followed by paracetamol (2 g/kg b.w.p.o) on the last day.
- *Group X:* Rats in this group the aqueous extract of *P. Dulce* (500 mg/kg b.w.p.o) were given for seven consecutive days followed by paracetamol (2 g/kg b.w.p.o) on the last day.

At the end of the experiment, blood sample were collected by puncturing retro-orbital plexus. Collected blood samples were centrifuged for separation of serum. The serum was separated and processed for estimation of biochemical parameters like serum creatinine, blood urea level and albumin.

Biochemical Estimations and Histopathological Studies

On 7th day, blood was withdrawn from retro-orbital plexus and serum was separated. The serum and urea samples were used for biochemical estimations like serum creatinine, urea, and albumin. All biochemical analysis was conducted using standard test kits. These kits were used as per the directions given along with it; Smart 5 Semi auto Biochemistry analyzer was used for the analysis. After end of the study, the animals from each group were sacrificed and kidneys were isolated. The rat kidney identified and carefully dissected for histopathological examination. All the preserved organs/tissues samples such as skin from all the groups were processed routinely and embedded in paraffin. The sections of 3-5 µ thickness were cut and stained with hematoxylin-eosin stain and observed under 40x. Histopathology examinations of all the thin sections were examined carried out and noted down.

Estimation of Serum Creatinine: Creatinine levels were estimated using the standardized kits which followed the Jaffe's reaction. Creatinine reacts with alkaline picrate (in ratio of 1:1) a colored creatinine picrate complex produces reddish color. The absorbance of the color complex is directly proportional to creatinine concentration in sample. It was measured at 492 nm (480-520 nm).

Estimation of Urea: Urea reacts with urease GLDH reagent and then urea decomposes to form ammonia, which further reduces to form NAD^+. The rate of formation of NAD^+ is directly proportional to the amount of urea present in the sample and it is estimated by monitoring absorbance change within fixed time at 340 nm wavelength

Estimation of Serum Albumin: Albumin reacts with a dye bromocresol green in a buffered medium to form a green colored complex. The intensity of the color formed is directly proportional to the amount of albumin present in the sample. The absorbance of sample was measured at 546 nm.

Statistical Analysis

The experimental data was obtained from the biochemical estimations were expressed as mean ± SEM for each group. The statistical analysis was conducted using one-way analysis of variance (ANOVA) followed by Dunnette's multiple comparison tests. Values $p > 0.05$ were considered non-significant, $p < 0.05$ as significant, $p < 0.01$ as highly significant and $p < 0.001$ as very highly significant respectively. All experimental data was computed for statistical analysis by using graph pad prism software.

Results And Discussion

Phytochemical Screening: Phytochemical screening results showed the presence of carbohydrates, flavonoids, proteins, phenol, steroid and tannin.

Acute Toxicity Studies: Acute oral toxicity for methanolic and aqueous extracts of *P. dulce* Roxb. bark using wistar rats, according to the OECD Guidelines, 423, was found to be in GHS Category 5, > 2000–5000 mg/kg body weight, with a LD_{50} cut off at 5000 mg/kg body weight. Animal did not cause any mortality during the study. All the animals showed no clinical signs of toxicity immediately after dosing and appeared normal up to four hours and showed no clinical signs of intoxication at daily observations up to 14 days. As the oral LD_{50} cut off value of the extracts of was found to be safe for use.

Biochemical Estimations

cisplatin (CP), gentamicin (GM) and paracetamol (PC) treated rats developed polyuric acute renal failure as assessed by measuring different biochemical parameters like blood urea, plasma creatinine and plasma albumin. The blood urea level in CP, GM, and PC treated rats increased significantly (***P < 0.001) compared with the vehicle treated group. Plasma creatinine levels were also increased very highly significantly in CP, GM, and PC treated rats. CP and GM treatment plasma albumin level increased significantly, and PC treated animals were having significantly (*P < 0.05) changes compared with vehicle treated animals. (Table 1).

After treatment with the methanolic extract, values of urea, creatinine and albumin were reduced significantly was observed in CP-treated animals. In aqueous treated group values for urea, creatinine and albumin were decreased significantly as compared to disease control but it seems less effective than methanolic extract (Figs. 1–3).

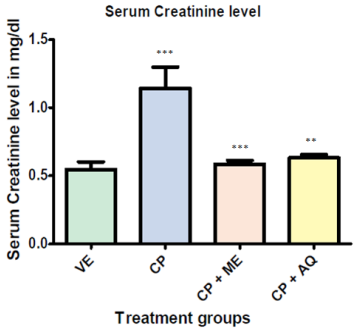
After treatment with the methanolic extract values of urea, creatinine and albumin were reduced significantly was observed in GM-treated animals. In aqueous treated group values for urea, creatinine and albumin were decreased significantly as compared to disease control but it seems less effective than methanolic extract (Fig. 4–6).

After treatment with the methanolic extract, values of urea, creatinine and albumin were reduced significantly was observed in PC-treated animals. In aqueous treated group values for urea, creatinine and albumin were decreased significantly as compared to disease control, but it seems less effective than methanolic extract (Fig. 7–9).

Table 1: Effect of administration of methanolic and aqueous extracts of *P. Dulce* bark on various biochemical parameters in cisplatin (CP), gentamicin (GT) and paracetamol (PC) induced renal damage.

Sr. No	*Group*	*Urea (mg/dL)*	*Creatinine (mg/dL)*	*Albumin (g/dL)*
I	Vehicle	38.342 ± 2.139	0.546 ± 0.140	2.900 ± 0.051
II	Disease Control- Cisplatin	72.392 ± 3.606***	1.142 ± 0.388***	3.855 ± 0.064 ***
III	Cisplatin + Methanol extract	51.217 ± 3.324 ***	0.586 ± 0.067 ***	2.978 ± 0.154 ***
IV	Cisplatin + Aqueous extract	63.713 ± 2.839 ***	0.632 ± 0.059 **	3.612 ± 0.062 ***
V	Disease Control -Gentamicin	144.601 ± 9.238 ***	1.897 ± 0.341 ***	3.815 ± 0.045 ***
VI	Gentamicin + Methanol extract	57.875 ± 4.186 ***	0.735 ± 0.098 ***	2.820 ± 0.055***
VII	Gentamicin + Aqueous extract	82.939 ± 5.664 ***	0.995 ± 0.145 ***	3.213 ± 0.046 ***
VIII	Disease control - Paracetamol	76.343 ± 5.779 ***	1.152 ± 0.396 ***	2.978 ± 0.050 *
XI	Paracetamol+ Methanol extract	39.243 ± 3.312 ***	0.566 ± 0.074 ***	2.860 ± 0.034 ***
X	Paracetamol+ Aqueous extract	43.231 ± 3.656 ***	0.580 ± 0.076 ***	2.887 ± 0.042 **

Values are expressed as Mean ± S.E.M. (n=6). The disease control group is compared with vehicle group and the treated groups are compared with the disease control group by One way ANOVA test followed by Dunnette's multiple comparison test ***P < 0.001 **P < 0.01, *p < 0.05.



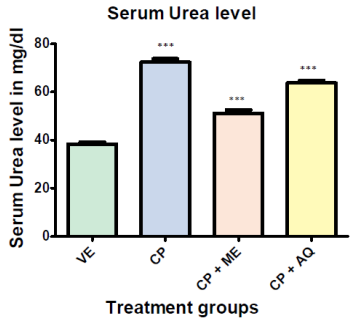


Fig. 1 & 2: Effect of *P. Dulce* on Serum Creatinine and Urea in Cisplatin induced nephrotoxicity in rats

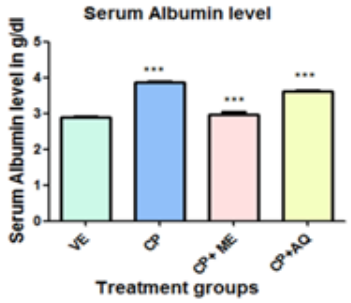
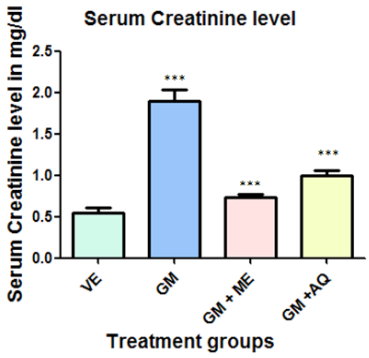


Fig. 3: Effect of *P. Dulce* on Serum Albumin in Cisplatin-induced nephrotoxicity in rats.



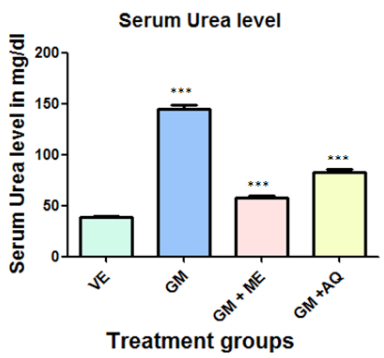


Fig. 4 & 5: Effect of *P. Dulce* on Serum Creatinine and Urea in Gentamicin induced nephrotoxicity in rats

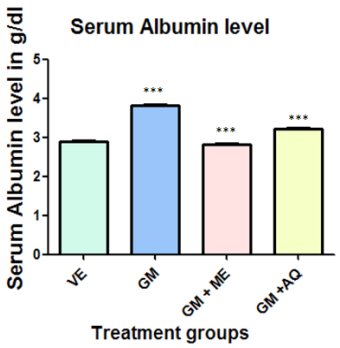


Fig.6: Effect of *P. Dulce* on Serum Albumin in Gentamicin induced nephrotoxicity in rats

Body Weight of Animals

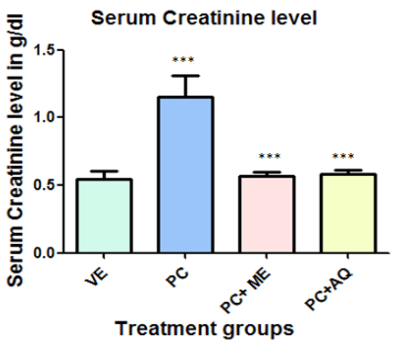
Animals were measured for their body weights at day 1 and just before sacrifice. Bodyweight was increased but not significantly observed in all the animals. In groups II and V body weight gained was not considered as compared to the vehicle control group (Table 2).

Histopathological Studies

Microscopic examination of the kidney showed the highest toxicity in diseased groups induced with cisplatin and gentamicin compared to paracetamol. At the same time, it was found that methanolic extracts were found to have more significant recovery as compared to aqueous extract. The aqueous extract was also significantly improved the pathological condition of the kidney. The histopathological changes in each kidney were observed for changes in glomerular membrane, endothelial tissue, interstitial congestion, tubular hemorrhage, inflammatory cell infiltrates and degeneration of cells.

Discussion

Cisplatin is currently used as one of the most important drugs in the treatment of a wide range of solid tumors of the head, ovary, and lung cancer. Gentamicin is used to gram-negative bacterial infections such as meningitis, lung, bone, joint and urinary tract infections.[15] Paracetamol treats to acute pain, aches and reduce fever.[16] Cisplatin gets accumulated in the tubular epithelial cells of the proximal kidney tubule, thus specifically causes nephrotoxicity. Gentamicin induces nephrotoxicity by inhibiting protein synthesis in renal cells, leading to necrosis of cells in the proximal renal tubule, resulting in acute necrosis, followed by acute renal failure. *Renal toxicity* in *paracetamol* is followed by reductions in glomerular filtration rate and cellular injury confined to the proximal tubule. CP, GM & PC induced renal impairment is evidenced by a very highly significantly ($p < 0.001$) increase in blood urea, serum creatinine, and serum albumin as well as acute necrosis that was evidenced through the histopathological examination of the kidney due to administration of cisplatin, gentamicin, and paracetamol in the regimen of nephroprotective effects. After treatment with the methanolic extract, values reduced significantly in CP, GM & PC treated animals. In the aqueous treated group, urea, creatinine, and albumin values decreased significantly compared to disease control, but it seems less effective than methanolic extract.



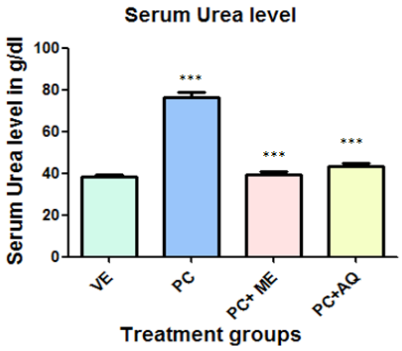


Fig. 7 and 8: Effect of *P. Dulce* on Serum Creatinine and Urea in Paracetamol induced nephrotoxicity in rats.

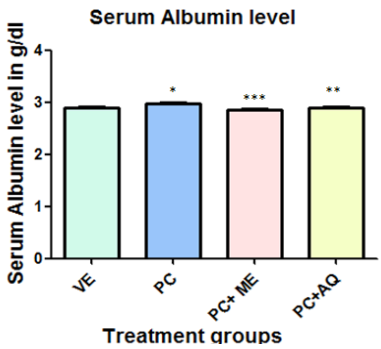
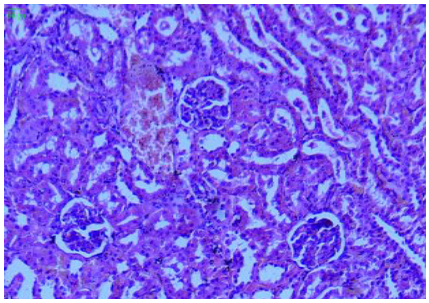


Fig.9: Effect of *P. Dulce* on Serum Albumin in Paracetamol induced nephrotoxicity in rats.

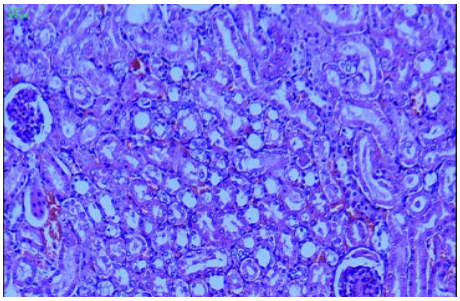
Table 2: Effect of administration of methanolic and aqueous extracts of *P. Dulce* bark on body weight in induced renal damage.

Sr. no	*Group*	*Day 0 (gm)*	*On day of scarification (gm)*
I	Vehicle	190.25 ± 4.27	197.92 ± 4.14
II	Disease Control- Cisplatin	188.08 ± 4.57	194.42 ± 4.55
III	Cisplatin + Methanol extract	188.33 ± 4.76	195.33 ± 5.13
IV	Cisplatin + Aq extract	188.83 ± 4.46	195.25 ± 4.06
V	Disease Control -Gentamicin	189.00 ± 4.80	194.08 ± 4.60
VI	Gentamicin + Methanol extract	188.67 ± 4.52	194.92 ± 4.50
VII	Gentamicin + Aq extract	188.92 ± 4.83	195.25 ± 5.15
VIII	Disease control - Paracetamol	189.33 ± 4.80	198.00 ± 4.84
XI	Paracetamol+ Methanol extract	188.83 ± 5.57	197.42 ± 5.94
X	Paracetamol+ Aq extract	188.67 ± 5.70	197.58 ± 5.54

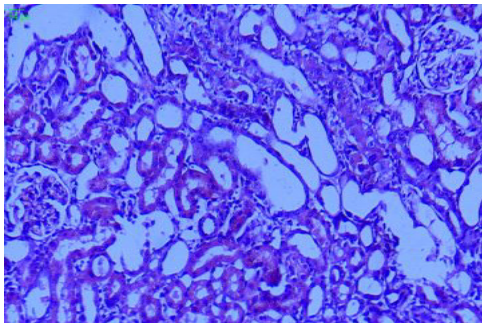
Values are expressed as Mean ± S.E.M. (n=6). The Disease control group is compared with vehicle group, and the treated groups are compared with the Disease control group by One way ANOVA test followed by Dunnette's multiple comparison test ***P < 0.001 **P < 0.01, *P < 0.05.



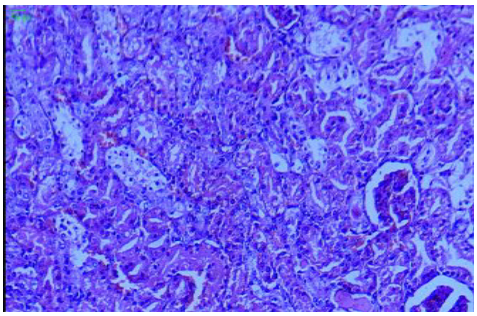
Group I: Vehicle- Saline
Normal Nephrons were observed, no any pathological changes were observed



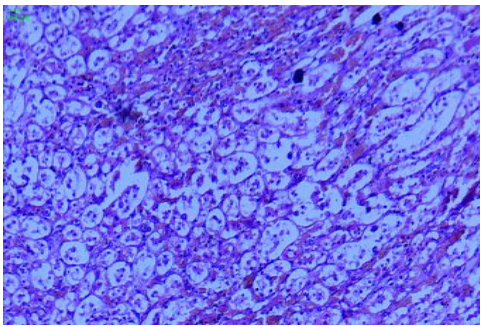
Group II: Disease Control- Cisplatin
Moderate inflammation and thickened basal membrane, loss of endothelial tissue, higher glomerular fibrosis, and tissue necrosis was observe



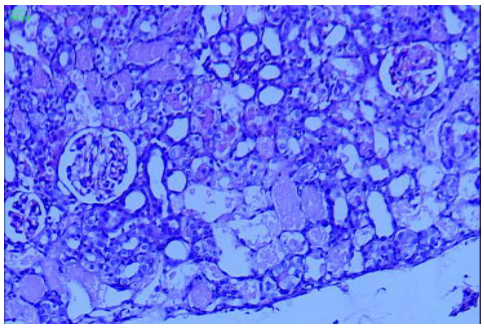
Group III: Protective group Cisplatin + Methanol extract
Mild inflammation of nephrons along with mild necrosis, no damage to glomerular membrane, mild inflammation, but no hemorrhage was observed



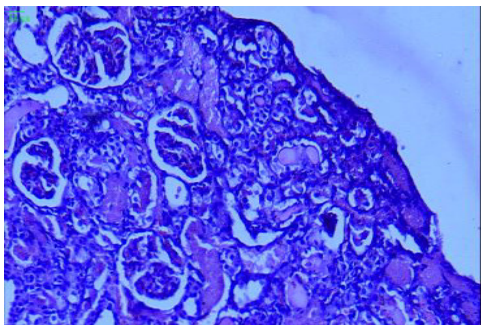
Group IV: Protective group- Cisplatin + Aq extract
Inflammation and tissue necrosis was moderate, whereas some endothelial loss was observed. Mild fibrosis and hemorrhage was observed



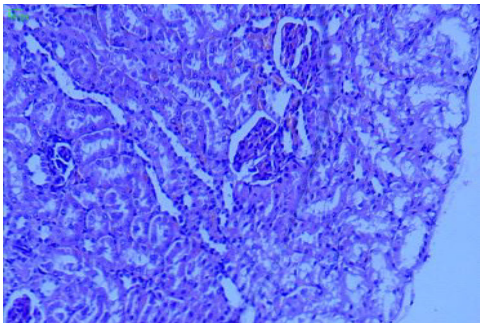
Group V: Disease Control -Gentamicin
Severe Inflammation and thickened basal membrane, moderate haemorrhage, severe endothelial swelling and disruption along with loss of endothelial tissue, higher glomerular fibrosis and tissue necrosis was observe



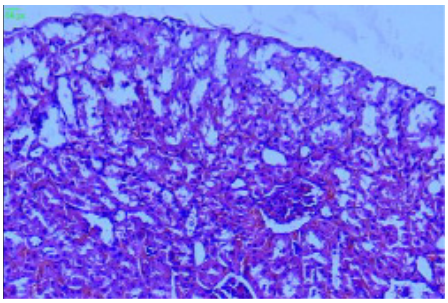
Group VI: Protective group - Gentamicin + Methanol extract
Mild inflammation of nephrons along with mild necrosis, no damage to glomerular membrane, mild inflammation but no haemorrhage was observed



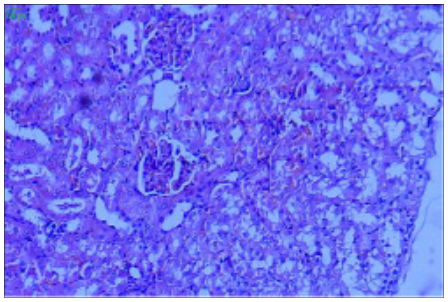
Group VII: Protective group- Gentamicin + Aq extract
Inflammation and tissue necrosis was moderate, whereas some endothelial loss was observed. Mild fibrosis and haemorrhage was observed



Group VIII: Disease control - Paracetamol
Moderate Inflammation and haemorrhage, Severe Endothelial swelling and disruption along with loss of endothelial tissue, moderate glomerular fibrosis in animals and no tissue necrosis was observed



Group XI: Protective group- Paracetamol+ Methanol extract
Mild inflammation and no haemorrhage were observed. No loss of endothelium but moderate tissue fibrosis in some of the animals was observed



Group X: Protective group- Paracetamol +Aq extract
Moderate Inflammation and mild haemorrhage, slight endothelial swelling and disruption along with loss of endothelial tissue, moderate glomerular fibrosis and mild tissue necrosis was observed.

The methanolic extract was found to have a more significant recovery as compared to aqueous extract. The aqueous extract was also significantly improved the pathological condition of the kidney. The histopathological changes in each kidney were observed for changes in the glomerular membrane, endothelial tissue, interstitial congestion, tubular hemorrhage, inflammatory cell infiltrates, and degeneration of cells. The bark of *P. dulce* exhibited good nephroprotective activity due to various chemical constituents such as tannins, flavonoids, steroids, and phenols containing drugs.

References

1. Hussain T, Gupta RK, Sweety K, Eswaran B, Vijayakumar M and Rao CV. Nephroprotective activity of *Solanum xanthocarpum* fruit extract against gentamicininduced nephrotoxicity and renal dysfunction in experimental rodents. Asian Pacific Journal of Tropical Medicine. 2012;5:68691. Available from: DOI: 10.1016/S1995-7645(12)60107-2
2. Fioretto P, Steffes W, Sutherland E, Goetz C, Mauer M. Reversal of Lesions of Diabetic-II. National Diabetes Information Clearinghouse. Diabetes Control and Complications Trial, U. S. Government Printing Office, NIH publication, Washington D.C, 1994;97: 3874-3875. Available from:DOI: 10.1056/NEJM199807093390202
3. Barbosa J, Steffes W, Sutherland E, Connett E, Rao V. Effect Of Glycemic Control on Early Diabetic Renal Lesions. Journal of American Medical Association. 1994;272(8):600-606. Available from: PMID: 8057515
4. Becker J, Perkovic V, Hewitson T. Pharmacological intervention in renal fibrosis and vascular sclerosis. Journal of Nephrology. 2001; 14:332-339. Available from:PMID: 11730265
5. Nagmoti DM, Khatri DK, Juvekar PR, Juvekar AR. Antioxidant activity and free radical-scavenging potential of *Pithecellobium dulce* Benth seed extracts. Free Radicals and Antioxidants. 2012; 2(2):37–43. Available from :DOI: 10.5530/ax.2012.2.7
6. Pithayanukul P, Ruenraroengsak P, Bavovada R, Pakmanee N, Suttisri R, Saen-oon S. Inhibition of Naja kaouthia venom activities by plant polyphenols. Journal of Ethnopharmacology. 2005;97(3): 527-533. Available from:doi: 10.1016/j.jep.2004.12.013.
7. Zapesochnaya GG, Yarosh EA, Syanidze NV, Yarosh GI. Flavonoids of the leaves of *Pithecellobium dulce*. Khimiya Prirodnykh Soedineni. 1980;2:252-253. Available from: doi:000/890/000890138.
8. Singh L, Mamta S. Oxidative DNA damage protective activity and antioxidant and hepatoprotective potentials of Pithecellobium dulce plant. 2nd International Conference and Exhibition on Nutritional Science & Therapy. Courtyard by Marriott Philadelphia Downtown, USA. Journal of Nutrition & Food Sciences. 2013;15-17. Available from:doi:10.1016/j.fct.2009.01.034.
9. Shankar K, Maheshkumar K. Antioxidant and free radical scavenging activity of *Pithecellobium dulce* (Roxb.) Benth wood bark and leaves. Free radicals and antioxidants. 2012;2(3):47-57. Available from : doi.org/10.5530/ax.2012.3.7
10. Raghu P, Hari P, Venkatesh P, Venkata K, Sudheer B. Antidiabetic activity of bark extract of *Pithecellobium dulce* benth in alloxan-induced diabetic rats. Natural product: An Indian journal.2010; 6 (4):201-204.
11. Khandelwal KR. Practical Pharmacognosy. Pune: Nirali Prakashan; 2008; 19thed. 149-56, 157-159.
12. Annie S, Rajagopal P, Malini S. Effect of *cassia auriculata* linn. Root extract on cisplatin and gentamicin – induced renal injury. Phytomedicine. 2005; 12(8): 555-560. Available from: doi: 10.1016/j.phymed.2003.11.010.
13. Harlalka GV, Patil CR, Patil MR. Protective effect of *Kalanchoe pinnata* pers. (crassulaceae) on gentamicin-induced nephrotoxicity in rats. Indian Journal of Pharmacology. 2007; 39(4): 201-205. Available from :DOI: 10.4103/0253-7613.36540
14. Ngangbam d, sayeri d, rajesh d, subhalakshmi d, meena d, Sucheta Devi. Nephroprotective effect of *elsholtzia blanda* benth. In paracetamol induced toxicity in albino rats. International Journal of Recent Scientific Research. 2018; 9(7): 28186-28189. Available from: DOI: 10.24327/IJRSR
15. Gentamicin sulfate. The American Society of Health-System Pharmacists. 16 August 2015.
16. Warwick C. Paracetamol and fever management. Journal of the Royal Society for the Promotion Health. 2008; 128 (6): 320-323. Available from: doi.org/10.1177/1466424008092794

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