IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Human Journals
Research Article
June 2017 Vol.:9, Issue:3

Evaluation of Nephroprotective Activity of *Chrysanthemum parthenium* against Gentamycin Induced Nephrotoxicity in Wistar Rats

IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Akila CR*, Neelima S, Purushothaman M, Pradeep Kumar M

Vasavi Institute of Pharmaceutical Sciences, Kadapa—516247

Submission: 2 June 2017
Accepted: 7 June 2017
Published: 25 June 2017

Keywords: Nephroprotection; *Chrysanthemum parthenium*; Ethanol; Gentamycin

ABSTRACT

Objective: The present study evaluated the nephroprotective activity of ethanolic extract of *Chrysanthemum parthenium* (EECP) in experimentally-induced nephrotoxic rats. **Methods:** Nephroprotective activity of EECP was studied against gentamycin (80mg/kg i.p.)-induced nephrotoxicity rats. The following parameters were evaluated; serum biomarkers such as Blood Urea Nitrogen (BUN), Uric acid, Urea, Albumin, Creatinine and Total Protein (TP), urine parameters such as Uric acid, Urea and creatinine and tissue antioxidant levels such as Glutathione (GSH), Lipid Peroxidation (LPO) and catalase. **Results and conclusion:** The normal group did not exhibit increase in serum parameters, but gentamycin toxicant group showed significant increase in serum parameters such as Blood Urea Nitrogen (BUN), Uric acid, Urea, Albumin, creatinine, urine parameters such as Uric acid, Urea and creatinine and LPO, whereas GSH and TP levels were markedly reduced. EECP group, EECP low dose (250 mg/kg p.o.) and high dose (500 mg/kg p.o.) treated groups showed significant decrease in Blood Urea Nitrogen (BUN), Uric acid, Urea, Albumin, creatinine, urine parameters such as Uric acid, Urea and creatinine and LPO and increase in GSH and TP levels. Based on the study findings in serum marker enzyme levels, urine parameters and antioxidant parameters, it is concluded that EECP possesses nephroprotective activity.

INTRODUCTION:

More than one fifth of people over ages of 65 years have some degrees of kidney disease. Nephrotoxicity is one of the most common kidney problems and occurs when body is exposed to a drug or toxin [1]. A number of therapeutic agents can adversely affect the kidney resulting in acute renal failure, chronic interstitial nephritis and nephritic syndrome because there is an increasing number of potent therapeutic drugs like aminoglycoside antibiotics, NSAID's, chemotherapeutic agents have been added to the therapeutic arsenal in recent years [2]. Exposure to chemical reagents like ethylene glycol, carbon tetrachloride, sodium oxalate and heavy metals such as lead, mercury, cadmium and arsenic also induces nephrotoxicity [3]. Nephroprotective agents are the substances which possess protective activity against nephrotoxicity. Gentamicin, a typical aminoglycoside antibiotic is widely used in clinical practices for the treatment of life threatening gram-negative infections. Gentamicin-induced nephrotoxicity is characterized by direct tubular necrosis, without morphological changes in glomerular structures. It generates hydrogen peroxide in rat renal cortex mitochondria and can also enhance the generation of reactive oxygen species (ROS). Abnormal production of ROS may induce cellular injury and necrosis via several mechanisms including peroxidation of membrane lipids, protein denaturation and DNA damage.The alteration in kidney functions induced by lipid peroxidation is a proximal event in the injury cascade of gentamicin-mediated nephrotoxicity [4, 5].

Chrysanthemum parthenium belonging to the family Compositae (Asteraceae) is reported to possess various chemical constituents like sesquiterpene lactones, flavonoid glycosides, pinenes, volatile oils [6, 7]. *C. parthenium* is traditionally used for the treatment of fevers, migraine headaches, rheumatoid arthritis, stomach aches, infertility, and problems with menstruation and labor during childbirth [8, 9]. It has also been used in psoriasis, allergies, asthma, tinnitus, dizziness, nausea, and vomiting [10]. It has multiple pharmacologic properties, such as anticancer, anti-inflammatory, cardiotonic, antispasmodic and as an enema for worms, Ancient Greeks and early Europeans used *C. parthenium* to repel insects [11] and treat bites and stings [12].

It has been claimed to be useful in nephrotoxicity in indigenous system of medicine [13]. But no systemic pharmacological studies were reported in the literature. Hence in the present study, the ethanolic extract of flower of *C. parthenium* was evaluated for nephroprotective activity.

MATERIALS AND METHODS:

Collection of Plant Material:

Flowers of *Chrysanthemum parthenium* were collected from local market, Kadapa District, Andhra Pradesh.

Ethanolic extract of *Chrysanthemum parthenium*:

The dried flowers of *C. parthenium* were collected, cleaned, dried and powdered in a grinder mixer to obtain a coarse powder and then passed through 40 mesh sieve. The powdered drug was subjected to defatting with n-hexane by soxhlet apparatus for 2 days. The solvent was recovered from their extract by distillation under reduced pressure.The plant powder (marc) was air dried after defatting. Again it is packed in Soxhlet apparatus for ethanol extraction for 18 hrs until to get clear solution in siphon tube. The dried extract thus obtained was kept in a desiccator and was used for further experiments.

Preparation of dose:

Weighed quantity of ethanolic extract of *Chrysanthemum parthenium* (EECP) was suspended in distilled water using 1%w/v sodium carboxymethyl cellulose which was supplied by Himedia Laboratories Pvt. Ltd, Mumbai. Based on the earlier research, two doses (250 and 500 mg/kg/day) were selected [14]. In control animals, 1% w/v sodium carboxymethyl cellulose was served as a vehicle and administered orally. The experiments were conducted 1 h after the oral administration. In multiple-dose study, the animals daily received the suitable oral dose of the EECP for a period of 21 days.

Experimental animals and research protocol approval:

Healthy adult male wistar rats weighing between 150-200gm were used for the present study. The animals were housed in groups of six and maintained under standard conditions (27±2ºC, relative humidity 44 - 56% and light and dark cycles of 10 and 14 hours respectively) and fed with standard rat diet and purified drinking water ad libitum for 1 week before and during the experiments.All experiments and protocols described in present study were approved by the Institutional Animal Ethical Committee (IAEC) of P. Rami Reddy Memorial College of Pharmacy (1423/PO/a/04/CPCSEA/104/2015).

Assessment of nephroprotective activity:

The experimental animals were randomly divided into 5groups (n= 6) and treated for duration of 21 days. Nephrotoxicity was induced by administration of Gentamycin (80 mg/kg i.p). Ethanolic extract of *C. parthenium* was freshly suspended in CMC and administered to animals by oral feeding needle.

Group I: (Normal) received 1% CMC for 21 days

Group II: (Gentamycin control) received Gentamycin (80 mg/kg i.p.) for 21 days

Group III: (EECP standard) received EECP (500 mg/kg/day p.o.) for 21 days

Group IV & V: (Low & high dose) received EECP (250 & 500 mg/kg/day p.o.) and

Gentamycin (80 mg/kg i.p.) for 21 days

Collection of Blood and Urine Samples:

The blood samples were collected from the retro-orbital venous plexus of rats without any coagulant for the separation of serum, at the regular intervals of the treatment. After collecting the blood, they were kept for 1h at room temperature and serum was separated by centrifugation at 2000rpm for 15min. Serum is collected and assayed for Blood Urea Nitrogen (BUN), uric acid, urea, total protein (TP), Albumin and Creatinine [15] according to standard methods.

Urine was collected over 24 hours on the 21st day by keeping the test animals in metabolic cages. The volume of collected urine samples was measured followed by estimation of biochemical parameters, namely urine Creatinine, urine uric acid and urine urea [15].

RESULTS AND DISCUSSION:

Biochemical Parameters in serum:

Blood Urea Nitrogen& Uric acid: Gentamycin caused an increase in the levels of BUN and uric acid in control group when compared to normal group.The normal animals treated with high dose of extract (G-III,500 mg/kg) produced effects similar to normal group without any treatment (G-I) shown in table no.1

The groups (G-V) receiving EECP(500 mg/kg) showed a significant ($p<0.05$) decrease in serum BUN levels, whereas group (G-IV) receiving EECP (250 mg/kg) surprisingly showed no significance in decreasing the serum BUN levels when compared to control group (G-II).

Urea, Total Protein and Albumin: There was significant ($p<0.001$) increase in the given serum levels in control group (G-II) when compared with normal group (G-I) shown in table no.1. The group (G-IV) receiving EECP(250 mg/kg) showed a significant ($p<0.05$) decrease in serum levels, whereas group (G-V) receiving EECP (500 mg/kg) showed more significant ($p<0.01$) decrease in serum levels than group (G-IV) when compared to control group (G-II). This indicates that higher dose is more effective in decreasing in serum levels than lower dose.The normal animals treated with high dose of extract (G-III, 500 mg/kg) produced effects similar to normal group without any treatment (G-I).

Creatinine: The result showed the effect of EECP on serum creatinine in normal and experimental groups. There was significant ($p<0.001$) increase in serum creatinine levels in control group (G-II) when compared with normal group (G-I) shown in table no.1.The

GROUPS	BUN (mg/dl)	URIC ACID (mg/dl)	UREA (mg/dl)	TOTAL PROTEIN (g/dL)	ALBUMIN (g/dL)	CREATININE (mg/dl)
I	35.17±2.44	1.22±0.06	39.17±1.66	3.88±0.56	2.86±0.18	0.37±0.05
II	68.83±4.34***	2.05±0.13***	67.50±9.41***	8.31±0.17***	5.80±0.28***	0.80±0.04***
III	40.00±4.94*	1.30±0.08***	42.67±2.71***	6.63±0.50*	3.41±0.16***	0.45±0.02***
IV	65.33±5.69	1.23±0.03***	48.67±2.88*	6.78±0.24*	4.46±0.38**	0.60±0.05*
V	50.00±1.63*	1.33±0.18***	44.33±3.38***	6.45±0.35*	4.30±0.28**	0.51±0.06**

normal animals treated with high dose of extract (G-III, 500 mg/kg) produced effects similar to normal group without any treatment (G-I). The group (G-IV) receiving EECP (250 mg/kg) showed a significant ($p<0.05$) decrease in Creatinine levels but the group (G-V) receiving EECP (500 mg/kg) showed a significant ($p<0.01$) decrease in creatinine levels when compared to control group (G-II).

BIOCHEMICAL PARAMETERS IN SERUM:

Table no: 1Effect of EECP on serum renal markers

All values are shown in mean ±SEM and n=6. *** indicates $p<0.001$ when compared with normal group. *Indicates $p<0.05$, **indicates $p<0.01$, ***indicates $p<0.001$, when compared with control group. BUN = Blood Urea Nitrogen.

BIOCHEMICAL PARAMETERS IN URINE:

Table no.2: Effect of EECP on uric acid, urea and creatinine

GROUPS	Uric acid (mg/dl)	Urea (mg/dl)	Creatinine (mg/dl)
I	1.73	17.75	13.8
II	4.53	21.25	25.2
III	1.45	12.5	14.4
IV	2.65	15.5	18.9
V	2.16	13.75	16.7

BIOCHEMICAL PARAMETERS IN URINE:

Effect on uric acid, urea and creatinine:

The result showed the effect of EECP on activity of uric acid, urea and creatinine levels in urine in both normal and experimental groups. There was remarkable increase of all the three levels in urine in control group (G-II) when compared with normal group (G-I) shown in the

table no.2. The normal animals treated with high dose of extract (G-III, 500 mg/kg) produced effects similar to normal group without any treatment (G-I).The group (G-IV) treated with EECP at low dose(250 mg/kg) decrease in all the parameters levels in urine whereas the group (G-V) receiving EECP at high dose(500 mg/kg) showed greatest protection in uric acid, urea and creatinine levels in urine, when compared to control group (G-II).

Antioxidant Parameters:

A significant decrease in the levels of catalase and glutathione (GSH) was observed in the control group (G-II) when compared to the normal group (G-I). The group-III receiving standard drug had significant increase in the catalase and GSH levels when compared to the control group (G-II). The groups-IV and V treated with EECP (250 mg/kg and 500 mg/kg) also exhibited a significant ($p<0.001$) increase in the catalase& GSH levels, when compared to the control group (G-II) shown in table no.3. The normal animals (G-III) treated with high dose (500 mg/kg) of extract produced effects similar to that of normal animals (G-I).

A significant increase in the levels of MDA was observed in the control group (G-II) when compared to the normal group (G-I). The group-II receiving standard drug had significant decrease in the MDA levels when compared to the control group (G-II). The groups-IV and V treated with EECP (250 mg/kg and 500 mg/kg) also exhibited a significant ($p<0.001$) decrease in the MDA levels, when compared to the control group (G-II).

Antioxidant Values

Table no: 3 Effect of EECP on tissue antioxidant levels

GROUPS	CAT (H_2O_2consumed/ gram tissue)	GSH (µg of GSH/mg)	LPO (µM /mg)
I	4.95±0.09	4.55±0.49	4.76±0.12
II	3.11±0.06$^{###}$	3.18±0.06$^{###}$	8.26±0.16$^{###}$
III	4.68±0.06***	4.53±0.10***	4.767±0.17***
IV	4.25±0.09***	4.25±0.14*	6.13±0.07***
V	4.21±0.19***	4.28±0.14*	5.18±0.20***

All values are shown in mean ±SEM and n=6. *** indicates p<0.001 when compared with normal group. *Indicates p<0.05, **indicates p<0.01, ***indicates p<0.001, when compared with control group

CONCLUSION

Major adverse effect of different drugs is nephrotoxicity. So it is a drug induced disease. This toxicity has been induced because of the release of the oxidants in kidney. Thus damaging or destructing the nephrons which are the basic functional units of kidney. The present study throws light on the effect of the ethanolic extract of *Chrysanthemum parthenium* in reducing the nephrotoxic effect that has been induced by gentamicin which is a broad spectrum antibiotic used to treat many ailments. This study gives the idea that when we use the plant along with the gentamicin like antibiotics will reduce the incidence of nephrotoxicity. Further detailed scientific investigation of the plant will be helpful in treatment of drug induced toxicity.

REFERENCES

1. Porter GA and Bennett WM. Nephrotoxic acute renal failure due to common drugs American Journal of Physiology, 1981; 241(7): F1-F8.
2. Hoitsma AJ, Wetzels JF and Koene RA. Drug induced nephrotoxicity. Aetiology, clinical features and management, Drug Saf, 1991; 6 (2): 131-147.
3. Paller MS., Drug induced nephropathies Med Clin North Am,1990; 74 (4):909-917.
4. Chandragouda R. Patil, Ramchandra B. Jadhav, Pushparaj K. Singh, Sneha Mundad and Prabhakar R. Patil. Protective Effect of Oleanolic Acid on Gentamicin Induced Nephrotoxicity in Rats. Phytother. Res. 2010; 24: 33-37.
5. Chaware V. J. Protective Effect of the aqueous extract of *Phaseolus radiates* seeds on Gentamicin Induced Nephrotoxicity in Rats. IJRPBS 2012; 3: 73-5.
6. Anonymous Feverfew, Lawrence Review of Natural Products; (1994). 1-2.
7. Bohlmann F, Zdero C., Sesquiterpene lactones and other constituents from *Tanacetum parthenium*. Phytochemistry; (1982),21: 2543-49
8. Hobbs C. Feverfew. Tanacetum parthenium. Herbal Gram ; (1989),20: 267-70.
9.Hendriks H, Bos R, Woerdenbag H, The essential oil of *Tanacetum parthenium (L)*. Flavour and Fragrance J; (1996),11: 367-71.
10. Barry MB,.Toxic Nephropathies. The Kidneys, (2000),2: 3–67.
11. DePooter H, Vermeesch J, Schamp N., Essential oils of *Tanacetum vulgare* L and *Tanacetum parthenium* L. J Essent Oil Res; (1989), 1: 9-13.
12. Baranov D., Everything you need to know about feverfew and migraines. In: Bratman S, Kroll D. The Natural Pharmacist; (1999), 175.
13.Jouad H, Haloui M, Rhiouani H, EI Hilaly J, Eddouks M., Ethnobotanical survey of medicinal plants for the treatment of diabetis, cardiac and renal diseases in the North centre region of Morocco (Fez-Boulemane). Journal of Ethnopharmacology; (2001), 77: 175-182.
14. Fabricant DS, Farnsworth NR.,. The value of plants used in traditional medicine for drug discovery. Environ. Health Perspect. Suppl, (2001), (1)109:69–75

15. Sharma RK, Rajani GP, Sharma V, Komala N. Effect of ethanolic and aqueous extracts of *Bauhinia variegata* Linn. on gentamicin induced nephrotoxicity in rats. Indian Journal of Pharmaceutical Education and Research 2011; 45(2): 192-198.