



Assessment of Nephroprotective Potential of Ethanolic Extract *Premna Tomentosa* (Ept) in Alcohol Toxicity in Male Albino Rats

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ABSTRACT

The nephroprotective prospective of *Premna tomentosa* extract against Alcohol induced nephrotoxicity in rats was investigated in the present study. The characterization of ethanol extract of *Premna tomentosa* (EPT) was performed using standard phytochemical analysis. Male albino wistar rats 36 in numbers were divided into 6 groups including control, negative control, positive control and various doses of EPT Treated groups; Nephrotoxicity was induced by alcohol (1ml/100gm b.wt) in animals. Rats intoxicated with Alcohol were fed with 500, 750 mg/kg dose of EPT and Liv 52 (1ml/100gm b.wt) for 60 days. Results show that EPT (500mg/kg b.wt) had a significant effect against alcohol induced nephrotoxicity in rats than EPT (750mg/kg b.wt) in blood parameters and consonantly good histopathological changes in kidney. The deleterious histopathological alterations in kidney associated with glomerular and tubular changes in alcohol intoxicated rats was evident. This result shows *Premna tomentosa* may be used as supplementary drug for alcoholics.



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INTRODUCTION

Alcohol prompted organ pathogenesis is due to the aggregation of toxic substances generated during alcohol metabolism (Mani *et al.*, 2016), which in turn generates reactive oxygen species (ROS) and other free radicals (Moncada *et al.*, 1994). It causes covalent alteration of cellular macro molecules, tissue

damage and kidney dysfunction (Offor *et al.*, 2019);

(Mahmoud *et al.*, 2015). Recent reports document that alcoholism remains a risk factor for post infectious glomerulonephritis, kidney graft failure acute kidney injury by initiating and promoting atherogenic risk factors, such as hyperuricemia, high blood pressure (Hebbani *et al.*, 2015) and diabetes (Schaffner and Ritz, 2012). Prevalence of renal disease is currently accretion at startling ratio (Adikwu and Bokolo, 2018). Acute renal injury happens when suddenly kidney loss the ability to excrete wastes, conserve electrolytes, concentrate urine and maintains fluid balance (Komail and Babu, 2018). Use of herbal products and supplements has increased tremendously over the past 3 decades with more than 80% of the world's population relies on use of traditional herbal remedies for primary health care (Shanmugam *et al.*, 2010). *Premna tomentosa* is a less known medicinal plant of high medicinal value (Narayan and Muthana, 1953). It is used to

treat different diseases such as diarrhea, headache, fever, conjunctivitis, skin diseases epilepsy and is

delineated to have antidiabetic, antidiarrheal, anti microbial, antioxidant, anti-inflammatory (Alam *et al.*, 1993), anthelmintic, antispasmodics (Banu *et al.*, 2015) and immunomodulatory activities (Devi *et al.*, 2003). Aqueous extract of *Premna tomentosa* leaves has been extensively used for the treatment of splenomegaly (Devi *et al.*, 2004). The water boiled with its leaf is used to treat human paralysis (Srinivas *et al.*, 2005). Additional analysis has enunciated the attainable benefit of *Premna tomentosa* in animal model of xenobiotic induced hepatotoxicity (Devi *et al.*, 1998). Furthermore investigations have shown that it contains more of phytochemical constituents which include alkaloids, terpenoids, flavonoids, steroids and phenolic compounds (Priyadarshini *et al.*, 2019). The high medicinal value of *Premna tomentosa*, our attentions were aroused to carry out this study to find the protective effect of ethanolic leaf extract of *Premna tomentosa* in alcohol induced nephrotoxicity in albino rats.

MATERIALS AND METHODS

Plant Material

The leaves of *Premna tomentosa* were collected from in and around Salem, Tamilnadu, India. The authentication of the plant was done by Dr. S. Sankaranarayana, Head, Department of Botany, Govt. Siddha Medical College, Anna Arch, Chennai with identification voucher No. GSMS/MB-Voucher Specimen No.23/2017.

Preparation of Plant Extract

The leaves were washed and light shaded dry for two weeks, after which was powdered and stored. The dried powder was extracted sequentially by ethanolic in hot continuous percolation method using Soxhlet apparatus. The solvent from the extracts was recuperated under reduced pressure using a rotary evaporator. The obtained crude ethanolic extract was stored at -20°C and is used for the experiments.

Animals

Study was carried out in accordance with guidelines and protocol approved by Institutional Animal Ethics Committee of Swamy Vivekanandha College of Pharmacy, India (SVCP/IAEC/PhD/2/02/2018). 36 Wistar albino rats (male) each weighing 150-200 g was obtained and were maintained on a 12h light/dark cycle in a temperature regulated (24±2°C) room. The rats were fed with standard pellets diet and water *ad libitum* throughout the experimental duration.

Experimental Design

The animals were divided into 6 groups (6 each) as follows;

Group I: Control Rats (1ml/100gm normal saline, b.wt).

Group II: Positive Control Rats (40% Alcohol 1ml/100g b.wt).

Group III: EPT (500mg/kg b.wt).

Group IV: Rats administered Liv 52 (1ml/100gm b.wt) along with 40% Alcohol 1ml/100g b.wt/day.

Group V: Rats administered EPT (500mg/kg b.wt) along with 40% Alcohol 1ml/100g b.wt/day.

Group VI: Rats administered EPT (750mg/kg b.wt) along with 40% Alcohol 1ml/100g b.wt/day.

The rats were feed using gavage method for 60 days. The blood samples were collected from retro orbital venous plexus and the serum is prepared through centrifuging at 10,000 rpm for 10 minutes at 30°C. Total protein, Albumin, Globulin, Urea, Creatine and Blood urea nitrogen were analyzed.

Histological analysis

For histopathologic studies, kidney was excised from the rats, and fixed in a neutral buffered solution of 10% formalin for 24h. The tissues were later embedded in paraffin and were thinly sectioned (5 microns) with a microtome and stained with hematoxylin and eosin. These sections were examined under digital microscope.

Statistical Analysis

Data were expressed in terms of Mean ± Standard Deviation (SD). One-way ANOVA followed by post hoc Duncan's multiple range test was used. The differences were considered significant at $p < 0.05$ by using SPSS 21 software.

RESULTS AND DISCUSSION

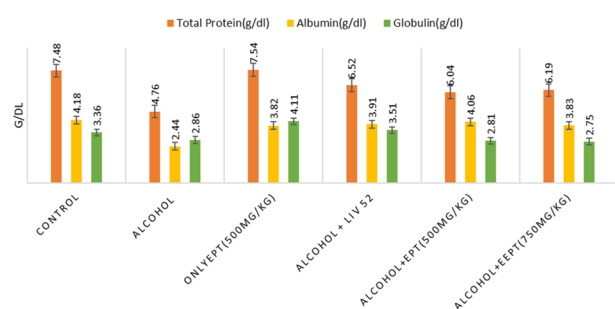
Graph 1 shows the effect of EPT on the levels of serum proteins, albumin and globulin of the control and experimental rats. EPT alone (Group III) treated rats haven't shown any statistically significant ($P < 0.05$) difference on levels of serum proteins as compared to that of control rats. However, significantly reduced levels of serum total proteins, albumin, and globulin was noticed in alcohol fed rats (Group II) as compared to the control rats, treated with EPT (Groups V and VI), the levels of serum proteins were elevated significantly as compared to the alcohol treated rats.

Table 1 shows the effect of EPT on renal function markers of control and experimental rats. EPT alone

Table 1: Effect of EPT and alcohol on serum urea, BUN and creatinine of control and experimental rats.

Groups	Total Urea (mg/dl)	BUN (mg/dl)	Creatine (mg/dl)
Control	26.45±1.28	12.32±0.65	0.67±0.09
Alcohol	54.7±2.20***	25.60±0.48**	1.31±0.30**
Only EPT (500mg/kg)	25.17±0.81	11.76±0.55	0.7±0.14
Alcohol + liv 52	44.63±1.17	20.70±0.66	0.68±0.07
Alcohol +EPT (500mg/kg)	36.5±2.54	17.18±2.06	0.68±0.07
Alcohol +EPT (750mg/kg)	38.58±1.44	18.15±0.80	0.62±0.04

Values are Mean±SD (n=6). Data for normal animal are considered as base line data; percentage increases is calculated with reference to normal control *p<0.05 versus control group,**p<0.001 versus control group,***p<0.001 versus control group.



Graph 1: Effect of EPT and alcohol on the serum proteins, albumin and globulin of control and experimental rats.

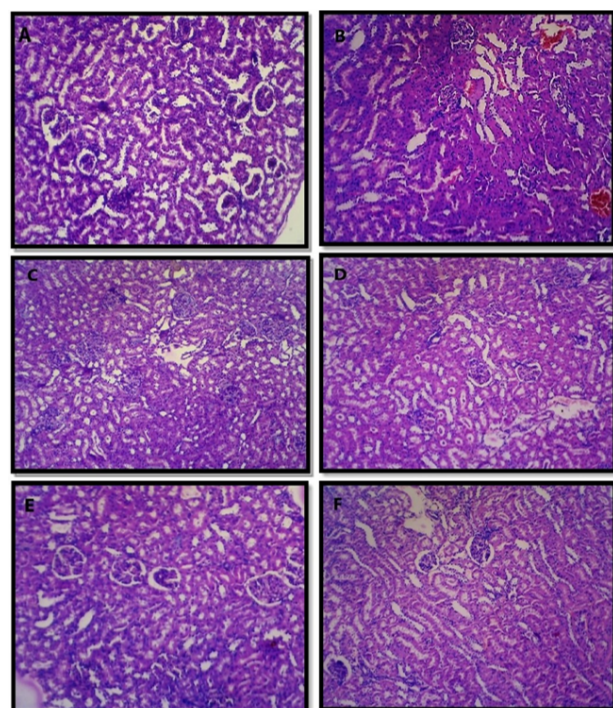


Figure 1: Evidence for the protective effect of *Premna tomentosa* in rats treated with alcohol (A) control (B) toxicant (C) EPT (D) Toxicated rats treated with Liv 52 (E) Toxicated rats treated with EPT 500 mg/kg p.o (F) Toxicated rats treated with EPT 750 mg/kg p.o.

(Group III) fed rats haven't showed any statistically significant ($P < 0.05$) difference on levels of serum urea, BUN and creatinine when compared with that of control rats.

The serum levels of urea, BUN and creatinine were remarkably increased ($P < 0.05$) in rats treated with alcohol alone (Group II) as compared to control and EPT alone treated rats. Supplementation with EPT (Groups V and VI) showed significantly decreased levels of serum urea, BUN and creatinine as compared to alcohol treated rats.

Histopathological section of kidney tissue shows normal architecture of glomerulus and tubules in control rats (Group I). Alcohol treated rats (Group II) kidney array the hypercellularity with reduced Bowman's space in the glomerulus and periglomerular inflammatory infiltration Figure 1B when compared to the normal cellularity and intact glomerular space in control group rats Figure 1A. The EPT alone at the dose of 500 mg/kg b.wt explains the normoarchitecture of glomerulus and tubules Figure 1C. Alcohol treated rats with Liv 52 (1ml/100gm b.wt p.o) states that slight increase in Bowman's space and reduced inflammatory infiltration Figure 1D. Administration of 750 mg/kg b.wt. Glomeruli shows mild focal segmental mesangial hypercellularity, Interstitium shows colloidal cast and there are no changes in tubules Figure 1F.

Alcoholic rats treated with of 500 mg/kg b.wt. Dose of EPT revealed merely normo architecture of the glomerulus and tubules Figure 1E. Treated rats with Liv 52 (1ml/100gm b.wt) shows near to the normoarchitecture but were comparatively in reduced form. Kidneys of rats fed with alcohol exhibit severe destructive changes in tubules, diffused cellular infiltration, and severe congestion of blood vessels. The rate of serum urea production exceeds the rate of clearance causes accumulation of serum urea which leads to nephrotoxicity (Alam et al., 2019).

Hence, serum urea concentration is generally

contemplated a more dependable kidney function prophecy than serum creatinine (Ravindra, 2010). Creatinine is a breakdown product of creatine and is removed from body by the kidneys. Increased urea and creatinine levels was considered as the sign of nephrotoxicity (Devi et al., 2016; Gutierrez et al., 2010) which is revealed in this study. Histopathological study reveals that EPT (500mg/kg) have nephroprotective potential on alcohol induced nephrotoxicity.

CONCLUSIONS

From the obtained results, it may be concluded that ethanol extract of *Premna tomentosa* manifests remarkable nephroprotective activity in alcohol induced nephrotoxicity in biochemical and also in attenuation of pathological changes in kidney tissues. It may due to the presence of therapeutic phyto constituents, antioxidant potential which reduces lipid peroxidation and oxidative stress.

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Conflict of Interest

The authors declare that they have no conflict of interest for this study.

REFERENCES

- Adikwu, E., Bokolo, B. 2018. Ethanolic leaf extract of *Ocimum gratissimum* ameliorates methotrexate-induced nephrotoxicity in albino rats. *Asian Journal of Medical Sciences*, 9(5):37–43.
- Alam, M., et al. 1993. Anti-inflammatory activity of *Premna tomentosa* Willd. in albino rats. *Ancient science of life*, 13(1-2):185–188.
- Alam, N., et al. 2019. Nephroprotective potential of Neermulli/Nerunjil Kudineer on Cisplatin induced nephrotoxicity in Wistar albino rats. *International Journal of Research in Pharmaceutical Sciences*, 10(3):1720–1729.
- Banu, J., et al. 2015. In vitro antioxidant activity of various extracts of aerial parts of *Premna tomentosa*. *International Journal of Chemical and Pharmaceutical Sciences*, 6(2):21–26.
- Devi, K. P., et al. 1998. Effect of *Premna tomentosa* on rat liver antioxidant defense system in acetaminophen-intoxicated rats. *Biomedical Research*, 19(5):339–342.
- Devi, K. P., et al. 2003. Immunomodulatory effects of *Premna tomentosa* extract against Cr (VI) induced toxicity in splenic lymphocytes—an in vitro study. *Biomedicine & Pharmacotherapy*, 57(2):105–108.
- Devi, K. P., et al. 2004. Assessment of the protective potential of *Premna tomentosa* (L. Verbenaceae) extract on lipid profile and lipid-metabolizing enzymes in acetaminophen-intoxicated rats. *Journal of Alternative and Complementary Medicine*, 10(3):540–546.
- Devi, M. A. R. L., et al. 2016. Evaluation of nephroprotective activity of ethanolic extract of *Annona reticulata* in gentamicin and cisplatin induced nephrotoxicity in rats. *Journal of Pharmaceutical Sciences and Research*, 8(9):995–1007.
- Gutierrez, R. M. P., et al. 2010. Nephroprotective activity of *Prosthechea michuacana* against cisplatin-induced acute renal failure in rats. *Journal of Medicinal Food*, 13(4):911–916.
- Hebbani, A. V., et al. 2015. Protective effect of aqueous bark extract of *Terminalia arjuna* against alcohol-induced hepato and nephrotoxicity in rats. *International Journal of Phytomedicine*, 7(2):142–153.
- Komail, M., Babu, A. N. 2018. Nephroprotective effect of *Jatropha curcas* fruit extracts against carbon tetrachloride induced nephrotoxicity in rats. *International Journal of Pharmacognosy and Phytochemical Research*, 9(7):943–946.
- Mahmoud, A. M., et al. 2015. *Ruta graveolens* and its active constituent rutin protect against diethylnitrosamine-induced nephrotoxicity through modulation of oxidative stress. *Journal of Applied Pharmaceutical Science*, 5(10):16–21.
- Mani, V., et al. 2016. Zingerone ameliorates hepatic and renal damage in alcohol-induced toxicity in experimental rats. *International Journal of Nutrition, Pharmacology, Neurological Diseases*, 6(3):125–132.
- Moncada, C., et al. 1994. Ethanol-derived immunoreactive species formed by free radical mechanisms. *Molecular Pharmacology*, 46(4):786–791.
- Narayan, V. L., Muthana, M. S. 1953. Essential oil from *Premna tomentosa*. *Journal of the Indian Institute of Science*, 35(1):55–67.
- Offor, C. E., et al. 2019. Protective effects of ethanol leaf-extract of *Cajanus cajan* (Linn) in alcohol induced nephrotoxicity in albino rats. *Trends in Telemedicine & E-health*, 2(1).
- Priyadarshini, G., et al. 2019. Preliminary phytochemical analysis of leaf extract of one medicinal plant *Premna tomentosa*. *International Journal of Pharmacy and Biological Sciences*, 9(1):271–274.
- Ravindra, P. 2010. Cisplatin induced histological changes in renal tissue of rat. *Journal of Cell and*

Animal Biology, 4(7):108-111.

Schaeffner, E., Ritz, E. 2012. Alcohol and kidney damage: a Janus faced relationship. *Kidney International*, 81(9):816-818.

Shanmugam, K. R., et al. 2010. Protective effect of ginger against alcohol-induced renal damage and antioxidant enzymes in male albino rats. *Indian Journal of Experimental Biology*, 48(2):143-149.

Srinivas, S., et al. 2005. Utility of serum prostate specific antigen doubling time (PSADT) in patients with androgen independent prostate cancer. *Journal of Clinical Oncology*, 23(16):4631.