https://ijrps.com

ISSN: 0975-7538
Research Article

Evaluation of hepatoprotective activity of *Ficus bengalensis*

Jagdish R. Baheti*[1], Ramesh K.Goyal[2]

[1]S.S.D.J. College of Pharmacy, Neminagar, Chandwad, Nasik, M.S, India
[2]Vice Chancellor, M.S. University, Vadodara, Gujarat, India

ABSTRACT

We have investigated the hepatoprotective activity of methanolic extract of bark of *Ficus bengalensis* against paracetamol and CCl_4 induced liver damage. Treatment of rats with paracetamol and CCl_4 produced a significant increase in the levels of Serum Glutamate Pyruvate Transaminase SGPT, Serum Glutamate Oxaloacetate Transaminase SGOT, Alkaline Phosphatase ALP, total and direct *bilirubin*. Rats pretreated with methanolic extract of barks of *F. bengalensis* 100 and 250 mg/kg body weight p.o. exhibited rise in the levels of these enzymes but it was significantly less as compared to those treated with paracetamol or CCl_4 alone. The results of methanolic extract of *F. bengalensis* were comparable with the standard hepatoprotective agent silymarin 100mg/kg. Maximum hepatoprotective effect was found to be at the dose of 250mg/kg body weight in case of CCl_4 induced hepatic damage while 500mg/kg body weight in case of paracetamol induced hepatic damage. Our data suggest that methanolic extract of *F. bengalensis* bark possesses a potential antihepatotoxic activity.

Keywords: *F. bengalensis;* Hepatoprotective; CCl_4; Paracetamol

INTRODUCTION

Ficus bengalensis Linn. Moraceae, known as Banyan in English and Bargad in Hindi is a large tree with spreading branches attaining height of 100 ft is well known to ancient Indian traditional medicine and grows almost everywhere in India (The Wealth of India, 1985). All the parts of the plant are acrid, sweetish and astringent to bowels. The leaves are indicated for ulcer. Its bark is antidiabetic and also used in piles, gonorrhea, syphilis, dysentery and inflammation of liver. The milky juice is aphrodisiac, tonic and anti-inflammatory (Kirtikar and Basu, 1988). Bengaloside, a glucoside (Augusti, 1975) and leucocynidine derivatives (Kumar & Augusti, 1989) isolated from bark have been reported to possess antidiabetic action and. Two flavonoid compounds dimethyl ether of leucopelargonidine 3-0 alpha L rhamnoside and 5,3 dimethyl ether of leucocyanidine 3-0 alpha L galactosyl celobioside have shown antioxidant effect in hyperlipidemic rats (Daniel et al., 1988).

Reactive oxygen species and free radicals play an important role in the etiology of various diseases such as inflammation, cataract, atherosclerosis, rheumatism, arthritis, ischemia reperfusion injury including liver disorders (Osawa et al., 1985). Paracetamol and CCl_4 share a common property of being converted into their respective reactive metabolites viz. N-acetyl p-benzoquinoneimine, NAPQI (Bessems, 2001) and halogenated free radical Packer et al., 1978 respectively by hepatic cytochrome P-450. Therefore, in the present study, the hepatoprotective effect of methanolic extract of *F. bengalensis* was evaluated in paracetamol and CCl_4 induced acute liver damage in the rats.

* Corresponding Author
Email: jbaheti@gmail.com
Contact: +91-9923380130 Fax: +91-2556-252029
Received on: 08-02-2011
Revised on: 15-03-2011
Accepted on: 18-07-2011

MATERIAL AND METHODS

CCl_4 was procured from E. Merck India Ltd. Mumbai; silymarin was obtained as gift sample from Cadila Pharma Ltd., India. Paracetamol was obtained as gift sample from Torrent Research Center, Ahmedabad. Standard kit of SGOT, SGPT ALP and bilirubin was obtained from Span Diagnostics Ltd. All other reagents used for the experiments were of analytical grade.

Preparation of methanolic extract of bark of *F. bengalensis*

The bark of *F. bengalensis* was collected from wildly grown plant and authenticated in our Pharmacognosy Department with the help of Botanist and a voucher specimen KB-PD 08/01 was preserved. It was air dried and powdered to 40 mesh and stored in airtight container till further use. 500 gm of the powder was defatted by petroleum ether and extracted with methanol using Soxhlet apparatus and the solvent was evaporated under reduced pressure. The methanolic extract was subjected to preliminary phytochemical testing for detection of major chemical groups (Harborne, 1998).

Animals

Wistar albino rats of either sex weighing between 150 and 200 g were used for the hepatoprotective study. The animals were housed in polypropylene cages and

maintained at 24±2°C under 12 hr light dark cycle and they were fed *ad libitum* with standard pellet diet and had free access to water. They were initially acclimatized for the study and the study protocol was approved by the Institutional Animal Ethics Committee as per the requirements of Committee for the Purpose of Control and Supervision on Animals CPCSEA, New Delhi.

Experimental protocol for hepatoprotective study

a. CCl4 induced hepatotoxicity

Rats were divided into five groups of five animals each. Group I served as vehicle control and received normal saline 5 ml/kg. Group II was administered with CCl_4 / Olive oil 1:1v/v, 0.7 ml/kg *i.p*.on alternate days (Singh et al., 1999). Group III and IV received methanolic extract 100 mg/kg and 250 mg/kg *p.o* respectively daily for seven days simultaneously with toxicant CCl_4/Olive oil. Group V was administered with reference drug, silymarin (Majumdar et al., 1998) 100mg/kg *p.o.* simultaneously with toxicant.

b. Paracetamol induced hepatotoxicity

In the paracetamol induced liver injury model, paracetamol 2 gm/kg p.o. suspension prepared using carboxy methyl cellulose was administered to all animals except the animals of the normal control group on 6th day (Packer et al., 1978). Silymarin 100 mg/kg p.o. was used as a standard. Group I, which served as normal control, received distilled water intraperitoneally. Group II received paracetamol 2 gm/kg p.o.single dose on 6th day. Group III received paracetamol 2 gm/kg *p.o*.single dose and silymarin 100 mg/kg p.o. simultaneously for 7 days. Group IV received paracetamol 2 gm/kg p.o. single dose and methanolic extract 100mg/kg p.o. simultaneously for same period. Group V received paracetamol 2 gm/kg *p.o*.single dose and methanolic extract 250 mg/kg p.o. simultaneously for same period.

Assessment of hepatoprtective activity

On the seventh day of the start of respective treatment the rats were anaesthetized by light ether anesthesia and the blood was withdrawn by making intracardiac puncture to the rats. It was allowed to coagulate for 30 minutes and serum was separated by centrifugation at 2500 rpm. The serum was used to estimate Serum Glutamate Pyruvate Transaminase, SGPT (Reitman, 1957), Serum Glutamate Oxaloacetate Transaminase, SGOT (Reitman, 1957) Alkaline Phosphatase, ALP (King, 1959). Total Bilirubin and Direct Bilirubin (Jendrassic,

Table 1: Effect of methanolic extract of *F. bengalensis* on CCl_4 and paracetamol induced hepatotoxicity

Group	Serum Biochemical Parameters				
	SGPT U/ml	SGOT U/ml	Alkaline Phosphatase K.A.Units	Total Bilirubin mg/dl	Direct Bilirubin mg/dl
NormalControl	31.70 ±4.56	38.95±7.51	12.99±0.45	0.87±0.04	0.16±0.09
CCl_4 0.7ml/kg	206.62±7.06[a]	197.58±6.10 [a]	39.82±1.54 [a]	2.59±0.42 [a]	0.851±0.20 [a]
MeOH ext 100mg/kg + CCl_4	91.37±4.24*	121.89±4.0*	22.13±0.08*	1.8±0.38*	0.28±0.08*
MeOH ext 250mg/kg + CCl_4	56.04±2.27**	89.63±7.74**	18.66±0.42**	1.19±0.2**	0.19±0.09**
Silymarin 100 mg/kg + CCl_4	44.95±5.51**	87.65±5.03**	16.25±0.15**	0.96±0.13**	0.25±0.09**
Paracetamol 2 gm/kg	254.38±17.13[a]	564.64±37.73[a]	45.58 ±0.43 [a]	2.78± 0.55[a]	0.92 ± 0.15[a]
MeOH ext 100 mg/kg + paracetamol	87.04±1.64[#]	138.15±3.62[#]	26.25±0.15[$]	1.59±0.13[#]	0.68±0.09[#]
MeOH ext 250 mg/kg + paracetamol	59.04± 3.40[$]	106.65±4.83[$]	19.18±0.37[$]	1.4±0.19[$]	0.61±0.01[$]
Silymarin 100 mg/kg + paracetamol	16.19±1.71[$]	48.31±2.96[$]	14.01±0.15[$]	0.99±0.1[$]	0.33±0.02[$]

[a] Statistically significant at p<0.001 when compared with normal control group

*Statistically significant at p<0.05 when compared with CCl_4 treated group

** Statistically significant at p<0.001 when compared with CCl_4 treated group

[#]Statistically significant at p<0.05 when compared with paracetamol treated group

[$]Statistically significant at p<0.001 when compared with paracetamol treated group

Values are mean ±S.E.M., n=5

1938).

The results of antihepatotoxic activity were presented as the mean±SEM of 5 animals each group. Results were analyzed statistically using analysis of variance ANOVA followed by Tukeys test. Values of *P<0.05* were considered significant.

RESULTS

Preliminary phytochemical screening indicated the presence of tannin flavonoids saponins and sugars. There was significant elevation of SGOT, SGPT, ALP and Bilirubin total and direct levels in the CCl_4 treated groups Table1. In groups orally treated with 100 mg/kg, 250 mg/kg of aqueous suspensions of methanolic extract of *F. bengalensis* and silymarin 100 mg/kg, above activities of enzymes were found to be significantly $p<0.001$ decreased as compared to the CCl_4 treated group. Maximum protection by methanolic extract of *F. bengalensis* against CCl_4 induced hepatic damage was offered at the dose of 250 mg/kg Table1.

Like CCl_4, paracetamol treated animals showed elevation of SGOT, SGPT, ALP and Bilirubin total and direct levels as compared to vehicle treated normal control group. In groups orally treated with 100 mg/kg and 250 mg/kg of aqueous suspension of methanolic extract and silymarin, above activities of enzymes were found to be significantly decreased $p<0.05$ as compared to the paracetamol treated control group.

DISCUSSION

Paracetamol and CCl_4 induced hepatic injuries are commonly used models for the screening of hepatoprotective drugs (Slater, 1965) and the extent of hepatic damage is assessed by the level of released cytoplasmic alkaline phosphatase and transaminases GOT and GPT in circulation (Osawa, 1990). The present investigation also revealed that the given dose of CCl_4 0.7 ml/kg, *i.p.* and paracetamol 2 gm/kg produced significant elevation in SGPT, SGOT and alkaline phosphatase levels indicating an impaired liver function. The massive production of reactive species may lead to depletion of protective physiological moieties glutathione and tocopherols etc. and ensuing widespread propagation of the alkylation as well as peroxidation, causing damage to the macromolecules in vital biomembranes. The investigation further reveals that the methanolic extract of *F. bengalensis* had been effective in offering protection, which is comparable to silymarin. The methanolic extract of *F. bengalensis* barks when administered to the rats exhibited protection against both paracetamol and CCl_4 induced liver injuries as manifested by the reduction in toxin mediated rise in serum enzymes. Both Paracetamol and CCl_4 share a common property of being converted into their respective reactive metabolites N-acetyl p-benzoquinoneimine and halogenated free radical by hepatic cytochrome P-450(Chenoweth, 1962).

Paracetamol, an analgesic and antipyretic agent is safe in recommended doses but produces hepatic necrosis when ingested in very large doses. It is established that at these relatively large doses paracetamol is biotransformed into a reactive metabolite N-acetyl p-benzoquinoneimine NAPQI by cytochrome P-450 mixed function oxidase(Packer et al., 1978). Paracetamol toxicity is enhanced by factors that cause GSH depletion, enhanced NAPQI formation, or reduction in the antioxidative capacity of the liver, it could be suggested that the partial hepatoprotection afforded by the methanolic extract may be ascribed to one or more of these factors.

Phytochemical investigations have revealed that barks of *F. bengalensis* shows presence of flavonoids, coumarinolignans and tannins. The literature has already documented the antioxidant and hepatoprotective value of flavonoid and coumarinolignans(Larson 1999). Thus, it appears that the hepatoprotection offered by *F. bengalensis* extract may be related to its free radical scavenging activity.

CONCLUSION

It is thus concluded that methanolic extract of *F. bengalensis* barks exhibited antihepatotoxic effect against paracetamol and CCl_4 induced hepatic damage. Further studies in progress in our laboratory for isolation and characterization of phytoconstituents may lead to development of lead nucleus for hepatic dysfunction.

REFERENCES

Augasti, K.T. 'Hypoglycemic action of bengalenoside, a glucoside isolated from *Ficus bengalensis* Linn. in normal and alloxan diabetic rabbits', Indian. J. Physiol. Pharmacol. Vol. 4, 1975 pp. 218-220.

Augusti, K.T., Daniel, R.S., Cherian, S., Sheela, C.G. and Nair, C.R. 'Effect of leucopelargonin derivative from *Ficus bengalensis* Linn. on diabetic dogs',. Indian J. Med. Res. Vol. 99, 1994 pp 82-86.

Bessems, J. G. & Vermuelen, N. P., 'Paracetamol /acetaminophen induced toxicity', Crit. Rev. Toxicol., Vol.31, 2001 pp.55-59.

Chenoweth, M. B. & Hake, C. L., The smaller halogenated aliphatic hydrocarbon, Annl Rev in Pharmacol, Vol.2, 1962 pp. 363-368.

Daniel, R.S., Mathew, B.C., Devi, K.S. and Augasti, K.T. 'Antioxidant effect of two flavonoids from the bark of *Ficus bengalensis* Linn. in hyperlipidemic rats'. Indian J. Expt. Biol. Vol.36, 1998 pp. 902-906.

Harborne., J B, Phytochemical Methods, 1998 pp. 52-58.

Jendrassik, L. & Grof, P., 'Method for determination of Bilirubin', Biochem, Vol.297, 1938 pp81-86.

King, E. J.,Method for determination of Alkaline Phosphatase, American J Clin Path, Vol.12, 1959 pp.85-89.

Kirtikar, K.R. and Basu, B.D. 'Indian Medicinal Plants', 3rd Edn, Vol III, International Book Distribution: Dehradun, 1988 pp. 2312-2313.

Kumar, K.R. and Augasti, K.T. 'Antidiabetic effect of leucocyanidin derivative isolated from bark of *Ficus bengalensis* Linn', Indian. J. Biochem. Biophys. Vol. 6, 1989 pp. 400-404.

Larson R. A, 'Antioxidants from higher plants', Phytochemistry, Vol.27, 1988 pp.969-978.

Majumdar A M, Anuradha S & Upadhye Pradhan A M,'Effect of *Azadirachta indica* leaf extract on carbontetrachloride induced hepatic damage in rats', Indian J of Pharma Sci, Vol.606, 1998 pp. 363-368.

Osawa, T. Kavakishi, S. & Namiki, M. 'Antimutagenesis and anticarcinogenesis mechanisms', Vol.II, 1990 pp. 139-144.

Packer, J. E., Slater, T. F. & Wilson, R. L, 'Reaction of CCl_4 related peroxy free radical with amino acids', Life Science, Vol.23, 1978 pp. 2617-2622.

Rege, N., Dahanukar, S. & Karandikar, S. M,'Hepatoprotetive effects of *Tinospora cordifolia* against carbon tetrachloride induced liver damage', Indian Drugs, Vol.2112, 1984 pp.544.

Reitman, A., 'Method for determination of SGPT and SGOT', American J Clin Path, Vol.28, 1957 pp. 56-60

Roy H Burdon, 'Free radical damage and its control', Elsevier Science B V. Netherland 1994 pp. 125-126.

Shukla, R., Gupta, S., Gambhir, J.K., Prabhu, K.M., Murthy, P.S. 'Antioxidant effect of aqueous extract of the bark of *Ficus bengalensis* in hypercholesterolaemic rabbits'. J. Ethnopharmacol. Vol.21, 2004 pp. 47-51.

Singh K, Khanna A K & Chander R,'Hepatoprotective activity of ellagic acid against carbon tetrachloride induced hepatotoxicity in rats', Indian J Expt Biol, 1999 Vol.37, 1025-1029.

Slater, T. F, Biochemical studies on chemical injury, 1965 pp. 10-15.

The Wealth of India. Raw Materials, Vol IV, F-G CSIR, Ministry of Natural Resources and Scientific Research: New Delhi; 1972 pp. 24-25.