

## EVALUATION OF HEPATOPROTECTIVE ACTIVITY OF *IMPATIENS GLANDULIFERA* AND *TRIUMFETTA BARTRAMIA*

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### Summary

*Impatiens glandulifera* is a tall annual herb 30 -60 cm tall and found in Assam, Arunachal Pradesh, Meghalaya, Manipur of Northeast India. *Triumfetta bartramia* is a erect herb upto 2 mt. in height and is distributed throught Northeast India. Studies are undertaken for evaluation of the hepatoprotective activity of the ethanolic extracts of indigenous medicinal plants like *Impatiens glandulifera* and *Triumfetta bartramia*. White healthy adult albino rats are used for experiment. Ethanolic extracts of *Impatiens glandulifera* and *Triumfetta bartramia* were evaluated for toxicity studies. Paracetamol in dose of 3mg /kg is used as necrosis causing agent and Silymarin in a dose of 100mg/kg is used as a standard drug .Evaluation of the hepatoprotective activity of the ethanolic extract of *Impatiens glandulifera* and *Triumfetta bartramia* were are done by estimating the serum level of the marker enzymes(SGOT and SGPT), serum bilirubin and total protein, where results were found to be significant. The Histopathological studies of the liver in *Impatiens glandulifera*, *Triumfetta bartramia* and *Silymarin* treated group shows that necrosis of the liver were reduced and shows good regenerative areas. Morphological studies shows that pre-treatment with extracts of *Impatiens glandulifera* and *Triumfetta bartramia* significantly reduce the liver weight of experimental animals as compared to positive control animals In conclusion, the present studies indicated that the ethanolic extracts of *Impatiens glandulifera* and *Triumfetta bartramia* possesses statistically significant hepatoprotective activity as compared with the standard(Silymarin).

**Keywords:** *Impatiens glandulifera*, *Triumfetta bartramia*, Hepatoprotective activity, Paracetamol

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### **Introduction**

Liver diseases remain one of the serious health problems. However we do not have satisfactory liver protective drugs in allopathic medical practice for serious liver disorders. Herbal drugs play a role in the management of various liver disorders. Most of which speeds up the natural healing process of the liver. Numerous medicinal plants and their formulations are used for liver disorders in ethno medical practice as well as traditional system of medicine. More than 15 plants are evaluated for the hepatoprotective action in the light of modern medicine (1).

The plants *Impatiens glandulifera* (family: Balsaminaceae) is annual herb 30-60cm tall, stem swollen at nodes, leaves simple opposite, alternate or whorled. It is distributed mainly in the northeast region of India and throughout the great Himalayan belt. Traditionally used for treating psychologically disturbed patient also used for treatment of joint pain, as emetic, and diuretic agent. The plant *Triumfetta bartramia* (family: Tiliaceae) is an erect herb 2mt in height, root system moderately deep and spreading, leaves ovoid, rhomboid or chordate. It is distributed in the tropical and subtropical India especially in the northeast region. Traditionally used as demulcent, in gonorrhoea, to facilitate child birth, bark and leaves are used in the treatment of diarrhoea, as diuretic etc.(2).

Silymarin is a flavanolignan that has been introduced fairly recently as hepatoprotective agent. Silymarin is a mixture of three structural component silibinin, silydianine, silychristine. Clinical trials have shown that Silymarin exerts hepatoprotective effects in acute viral hepatitis poisoning by ethanol paracetamol and CCl<sub>4</sub>. Many studies have shown the beneficial effects of Silymarin as Hepatoprotective agent.(3)

Not much work has been reported on the properties of this plant. Keeping in view the present study has been undertaken to investigate the hepatoprotective activity of *Impatiens glandulifera* and *Triumfetta bartramia* on paracetamol induced liver damage in rats.

### **Materials and methods**

#### ***Plant material:***

The plants *Impatiens glandulifera* and *Triumfetta bartramia* were collected from places in and around Dibrugarh University, Dibrugarh, Assam. The plant material was taxonomically identified by the taxonomist of Department of Life Sciences of Dibrugarh University and the voucher specimen is kept in the department for future reference.

***Extraction:***

The leaves of both the plants were dried under shade and then powdered with a mechanical grinder which was then subjected to successive extraction in a soxhlet apparatus using petroleum ether( to remove the fatty materials) and ethanol. The ethanol extract is concentrated under reduced pressure. The resulting ethanolic extract was then used for hepatoprotective studies.

***Experimental animals:***

Studies were carried out using male Wistar albino rats 150-200gm in weight. They were obtained from the animal house of the Department of Pharmaceutical Sciences of Dibrugarh University, Dibrugarh, Assam. The animals were maintained in standard laboratory conditions of temperature humidity and 12 hrs light and 12hrs dark cycles. They were fed with standard rat feed and water ad libitum (filter water).All animals were acclimatised to lab conditions for a week before commencement of the experiment.

***Drugs and Chemicals:***

All the drugs and chemicals used were of analytical grade.Silymarin was purchased from Centaur pharmaceuticals ,Goa,Paracetamol was purchased from Centaur pharmaceuticals ,Goa.SGOT, SGPT, ALP,bilirubin and Total protein kit were procured from Dade Behring Inc Newark,DE,USA.Ethanol from Bengal chemicals and pharmaceuticals, petroleum ether and methanol from Ranbaxy S.A.S.Nagar.

***Animal treatment:***

The animals were divided in five groups of eight animals each. Group1 served as normal control and given CMC at a concentration of 0.5% for 7 days.Group2 served as positive control Group-3 served as standard(silymarin is given in a dose of 100mg/kg) Group 4 served drug treatment of *Impatiens glandulifera* extract and group 5 served as drug treatment of *Triumfetta bartramia* extract(both in a dose of 250mg/kg)dissolved in 0.5%CMC.Acute liver damage was produced in the animals of Group 2,3,4 and 5 by oral administration of paracetamol(as a single dose of 3gm/kg body weight) on the 7<sup>th</sup> day.The biochemical parameters were estimated after 18 hrs of fasting following last dose.(4)

***Collection of blood sample:***

At the end of experimental period the animals are sacrificed after ether anesthesia, blood samples were collected by direct cardiac puncture and kept in EDTA containing vials for 10 minutes, then it is centrifuged for 15-20 minutes at 2000 rpm the serum was collected and used for analysis like biochemical estimations like Aspartate aminotransferase (AST) Alanine amino transferase (ALT), Total bilirubin (TB) and total protein (TP) as per standard procedures.(4,5,6)

**Statistical Analysis**

Results were presented as mean±S.E.M. for all values. One way ANOVA was used to statistically analysed the results followed by Students 't' test .The level of significance is kept at P<0.05

**Results**

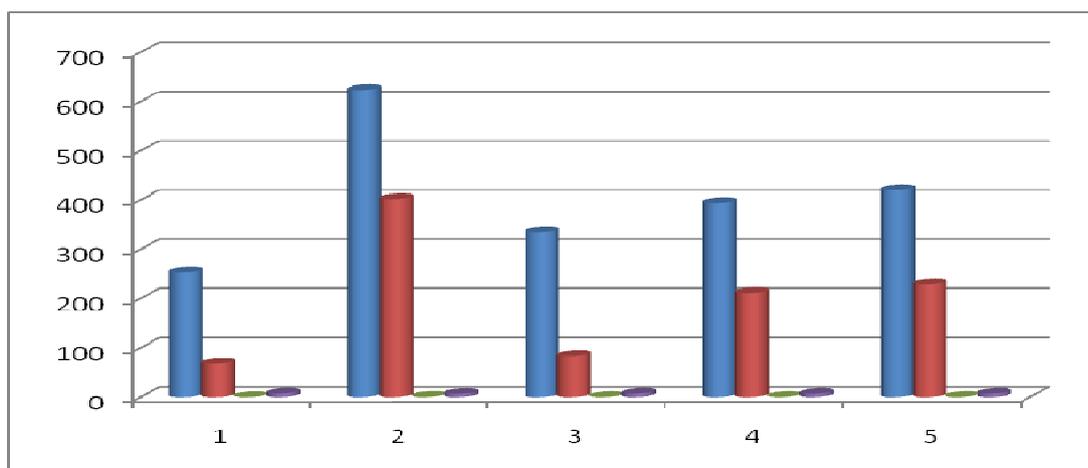
The level of AST and ALT is shown in the table this shows that the group2 animals shows marked increase in the levels of hepatic enzymes and therefore more liver damage while the test extract treated groups 4&5 shows significantly low levels of hepatic enzymes and are comparable with Silymarin treated group. It shows significant hepatoprotective activity.Ethanollic extracts of test material also shows statistically significant hepatoprotective activity as they reduced bilirubin and total protein to a level which can be compared with the Silymarin treated group:

Group	Treatment	AST (U/L)	ALT (U/L)	TB mg/dl	TP g/dl
Group1	control	253±0.47@	68±.18@	0.3±0.0072@	7.3±0.175@
Group2	+control	624±1.88#@	402±.47#@	0.7±0.137@#	6.6±0.065@#
Group3	standard	335±1.41#	84±.94#	0.4±0.011*	7.2±0.274*
Group4	Test extract1	394±1.03*#	211±.75*#	0.5±0.121*#	6.9±0.201*#
Group5	Test extract2	421±1.46*#	230±1.55*#	0.5±0.041*#	7.1±0.247*#

\*P<0.05 significantly different from positive control

# P<0.05 significantly different from Silymarin

@ P<0.05 significantly different from control



**Column graph showing comparison of selected parameters**

***Histopathological studies***

Histopathological studies on the liver sections are carried out to ascertain the effects of plant extracts and paracetamol on cytoarchitecture of the liver. Paracetamol treated groups shows various degree of pathological degenerative changes. Treatment with Silymarin, *Impatiens glandulifera* extract and *Triumfetta bartramia* extract showed good regenerative areas.

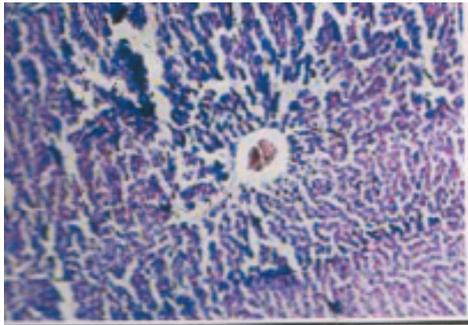


Fig **a**: Photomicrograph shows structure of a normal rat liver, H&E stain (X 5 X).

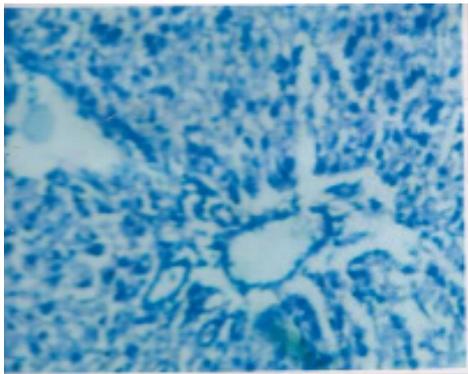


Fig **b**: Photomicrograph shows liver cell injury, caused by Paracetamol, H&E stain (X 50 X).

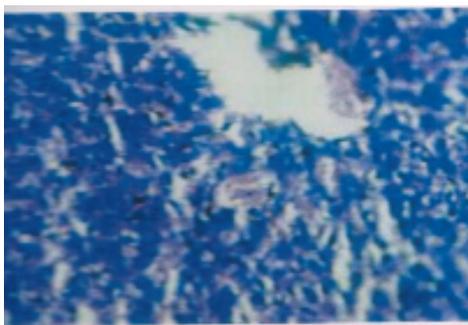


Fig **c**: Photomicrograph shows regenerative area of liver tissues by Silymarin, H&E stain (X 20 X).

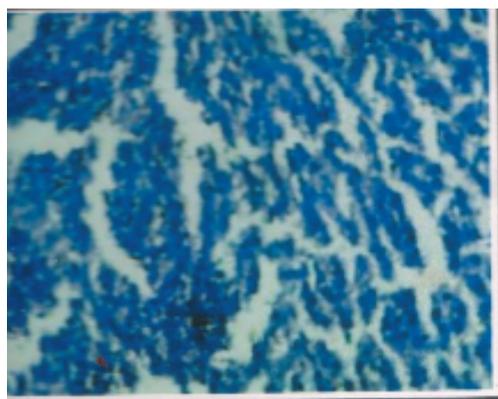


Fig. d: Photomicrograph shows regenerative area of liver tissues by *Impatiens glandulifera* Linn. H&E stain (X 20 X).

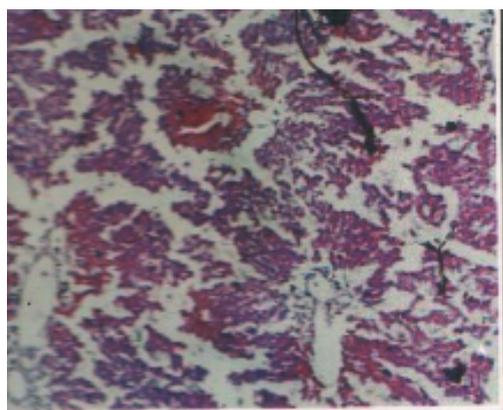


Fig. e: Photomicrograph shows regenerative area of liver tissues by *Triumfetta bartramia* Linn. H&E stain (X 5 X).

### Discussion

Paracetamol (acetaminophen) is a commonly and widely used analgesic and antipyretic agent. Hepatotoxic doses of acetaminophen deplete the normal levels of hepatic glutathione, when NAPQI covalently binds to cysteine groups on proteins to form 3-(cystein-S-yl) acetaminophen adducts(17). The glutathione protects hepatocytes by combining with the reactive metabolite of paracetamol thus preventing their covalent binding to liver proteins<sup>20</sup>. In living systems, liver is considered to be highly sensitive to toxic agents. The study of different enzyme activities such as SGOT, SGPT, total bilirubin and total protein have been found to be of great value in the assessment of clinical and experimental liver damage (18). In the present investigation it was observed that the animals treated with acetaminophen resulted in significant hepatic damage as shown by the elevated levels of serum markers. These changes in the marker levels will reflect in hepatic structural integrity. The rise in the SGOT is usually accompanied by an elevation in the levels of SGPT, which play a vital role in the conversion of amino acids to keto acids(19). The extracts of *Impatiens glandulifera* and *Triumfetta bartramia* when given to Group4 and 5 showed decrease levels of AST and ALT moreover when treated with the test extracts the rats of Group 4&5 also shows decreased level of total bilirubin and increase in total protein level which are found to be extremely high and low in the rats treated

with only paracetamol causing hyperbilirubinemia and hypoglobulinemia. This occurs in chronic liver diseases. The test extract treated groups also decrease the liver weight which increases during the liver toxicity. The histopathological study shows that in the *Impatiens glandulifera*, *Triumfetta bartramia* and silymarin treated groups the necrosis of liver were reduced to inflammatory cells. In conclusion, the present studies indicated that the ethanolic extracts of *Impatiens glandulifera* and *Triumfetta bartramia* possess statistically significant hepatoprotective activity as compared with Silymarin.

#### **Acknowledgement**

The authors would like to thank Dr. A. Bhattacharya, Head, Department of Pharmaceutical Sciences, Dibrugarh University, for providing all the necessary equipments and moral support. The authors also thank Dr. Islam of Department of Life Sciences of Dibrugarh University for the identification of the plant.

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