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Hepatoprotective Activity of *Boerhavia diffusa* against Paracetamol induced toxicity in rats

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ABSTRACT

The present study was designed to evaluate the hepato protective activity of Boerhavia diffusa against paracetamol induced hepatotoxic rats. Administration of paracetamol (3g/kg. b.wt.,) produced significant changes in the hepatocytes which was reflected in the altered parameters such as ALT, AST, GSH, Protein and Serum creatinine levels. Treatment with B.diffusa root extract (1g/kg. b.wt.,) produced remarkable changes and brought back the altered parameters to near normal, proving its hepato protective activity.

Keywords: Hepatotoxicity, Paracetamol, *Boerhavia diffusa*, Hepato protection .

INTRODUCTION

Liver is the largest and most complex internal organ in the body. It plays an important role in the maintenance of the internal environment through its multiple and diverse functions. It is involved in the intermediary metabolism of proteins, fats, and carbohydrates [1] and in the synthesis of the number of plasma proteins such as albumin, fibrinogen and clotting factors, in the production of various enzymes and formation and excretion of bile. It acts as a storage depot for proteins, glycogen, various vitamins and minerals. Hence, any injury to it or impairment of its function has grave implication on the health of the affected person. Every year about 18,000 people are reported to die due to liver cirhhosis caused by hepatitis [2].Although viral infection is one of the main reasons of hepatic injury, xenobiotics, excessive drug therapy, environmental pollutants and chronic alcohol ingestion can also cause severe liver injury.

Liver disease is still a world wide health problem. Unfortunately, conventional and synthetic drugs used in the treatment of liver disease are inadequate and sometimes can have serious side effects [3].

Boerhavia diffusa is a perennial diffuse herb, popularly known as mukkarattai in Tamil, spreading Hogweed in English, common name Punarnava and found throughout India as a weed in wastelands and roadsides [4].The roots are reported to be diuretic and laxative and are given for the treatment of jaundice. The roots have also been found to be anti-inflammatory [5], antifibrinolytic [6], Anticonvulsant, Hepato protective [7].A large number of compounds have been isolated from the roots of *B.diffusa* L.namely punarnavine, β-sitosterol, β-D glucoside tetracosanoic, hexacosanoic, stearic, palmitic, arachidonic acid, hentriacontance, ursolic acid and punarnava-voside [8].

Only a fewplants are really very promising hepato protective agents. Realizing the importance and common use of the roots of *B.diffusa* in the treatment of liver disorder by several tribes in India, it was decided to investigate the hepato protective activity of *B. diffusa*.

EXPERIMENTAL SECTION

Plant Material

Boerhavia diffusa roots were collected from Chidambaram in Cuddalore district of Tamilnadu, India in the month of March 2009 during the early hours of the days. The plant was identified and authenticated at the herbarium of Botany directorate, faculty of science. The roots were shade dried and powdered. The powdered roots were kept in airtight container in a deep freezer until the time of use.

Preparation of Extract

One hundred gram *Boerhavia diffusa* root powder was mixed with 1000 ml of distilled water and stirred magnetically overnight (12 h) at 37ºC.This was repeated three consecutive times. The residue was removed by filtration and the extract evaporated to dryness at a lower temperature (<40ºC) under reduced pressure in a rotary evaporator. The residual extract was dissolved in normal saline and used in the study. The yield of the extract was approximately 13.5g.

Animals

Male albino rats weighing about 120-180 g were obtained from the Indian Institute of science, Bangalore. The animals were housed in propylene cages and maintained in controlled temperature with 12 hours period of light and dark and fed with standard rat feed and water.

Induction of Liver damage

Liver damage was induced in rats by administering paracetamol at a dose of 3g/kg of body weight orally.

Experimental design

The animals were grouped as follows and each group consisted of 6 rats.
Group –I: The animals in group 1 served as control and received distilled water.
Group-II: The group 2 rats served as test and were administered with paracetamol at a dose of 3g/kg body weight orally.

Group-III: The animals in group 3 were treated orally with paracetamol (3g/kg body weight) and Boerhavia diffusa (1g/kg body weight).

After the completion of experimental regimen the rats were fasted over night and blood samples were collected by puncturing the retro orbital plexus under light ether anesthesia. Serum samples were used for the determination of various parameters.

Statistical analysis

The results are presented as mean ± SD. Data is statistically analysed using student's 't' test. P value lower than 0.05 was considered as statistically significant.

RESULTS AND DISCUSSION

Paracetamol (N-acetyl-P-amino phenol, acetaminophen) a widely used analgesic and antipyretic drug is known to cause hepatotoxicity in experimental animals and human at high doses. It is mainly metabolized in the liver to excretable glucuronide and sulphate conjugates. However, hepatotoxicity of paracetamol has been attributed to the formation of toxic metabolites. Over dose of paracetamol leads to mitochondrial dysfunction followed by acute hepatic necrosis.

All these events culminate in functional and morphological changes leading to loss of integrity of cell membranes which is evidenced by the rise in levels of serum marker enzymes . This occurs because of hepato cellular damage due to the reduced activity of the antioxidant enzymes and disturbance of Ca^{2+} homeostasis [9].

Table: 1 Effect of *Boerhavia diffusa* in various biochemical parameters

S.No	Groups	Particulars	AST (U/L)	ALT (U/L)	GSH (micromole/L)	Protein (g/dL)	Creatinine (mg/dL)
1.	I	Normal rats	38.10±7.62	38.12±7.62	11.7±1.4	6.42±0.072	1.661±0.020
2.	II	Paracetamol Induced rats	70.50±11.15*	70.33±11.50*	7.10±7.90*	3.74±0.064*	2.833±0.185*
3.	III	Paracetamol Induced + Herbal drug treated rats	36.12±7.72*	36.12±7.50*	10.10±0.90*	6.17±0.15*	1.888±0.125*

Values are mean ±Standard error (n=6). Group II compared with Group I; Group III compared with Group II. = significantly different at P<0.05*

Aminotransferases such as ALT and AST are liver specific enzymes and are considered to be very sensitive and reliable induces for measuring hepatotoxic as well as hepato protective effect of various compounds. Hepatic necrosis induced by paracetamol usually associated with elevated levels of serum enzymes that are indicative of cellular leakage and loss of functional integrity of the cell membrane in liver [10, 11]. In the present study, there was a rise in the levels of ALT and AST in paracetamol induced group (Table ,1). Toxicity of paracetamol was reduced by *Boerhavia diffusa*.

GSH is a major non-protein thiol in living organisms which plays a central role in coordinating the body's antioxidant defense process. GSH is a critical determinant of tissue susceptibility to

oxidative damage and the depletion of GSH has been shown to be associated with an enhanced toxicity to chemicals including paracetamol [12].Decline in GSH content in the serum of paracetamol intoxicated rats was increased in *Boerhavia diffusa* administered group.

The site specific oxidative damage of the susceptible amino acids of protein is now recorded as the major cause of metabolic dysfunction during pathogenesis [13]. The lowered level of total protein was observed in paracetamol treated group which was brought back to near normal in co-treatment of *B.diffusa*.

Creatinine is the end product of muscle catabolism which is removed by the liver in a constant rate. The serum creatinnie concentration is the most commonly used index for liver function. The level of creatinine in the blood rises if liver does not function properly [14].In the present study also, we observed the increased level of creatinine in paracetamol induced rats. Oral administration of *B.diffusa* restoned the level of creatinine in paracetamol treated rats.

The flavanoids present in *Boerhavia diffusa* may probably prevent the accumulation of excessive free radicals and protect the liver against paracetamol intoxication .Hepato protective activity of *Boerhavia diffusa* can have important chemical implications in the future treatment of liver disorders.

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