HEPATOPROTECTIVE ACTIVITY OF VARIOUS EXTRACTS OF *Cayratia carnosa* (Wall. Ex Wight) Gagnep. IN PARACETAMOL INDUCED HEPATOTOXICITY IN ALBINO RATS

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**ABSTRACT**

Objective: In the present study, the antihepatoxic activity of the various extracts of the whole plant of *Cayratia carnosa* was evaluated using paracetamol induced hepatotoxicity in albino rats.

Method: The degree of protection against liver toxicity was determined by measuring the serum biochemical parameters viz. SGPT (serum glutamic pyruvate transaminase), SGOT (serum glutamic oxalo acetate transaminase), ALP (serum alkaline phosphatase) and Bilirubin (Total, Direct and Indirect). In addition, histopathological examination of the liver was also studied. Silymarin at the dose of 25 mg/kg, p.o. was used as reference standard drug and it exhibited significant hepato-protection.

Result: Ethanolic extract at 400 mg/kg, which was comparable to that of standard drug silymarin while the aqueous extract 200 mg/kg treated animals exhibited the least hepato-protective activity.

Conclusion: The results obtained in this study provide a scientific support to the traditional folkloric use of *Cayratia carnosa* as a hepato-protective agent.

Keywords: *Cayratia carnosa*, Hepato-protective, Serum marker enzymes, Histopathology

**INTRODUCTION**

Liver is the most important organ concerned with metabolic activities[1]. It plays a major role in detoxification and excretion of many endogenous and exogenous compounds. Any injury to it or impairment of its functions may lead to implications on one’s health[2,3]. Living in the world of inadequately controlled environmental pollution and use of potent drugs has made the liver to get exposed to a variety of xenobiotics and therapeutic agents. Thus the disorders associated with this organ are numerous and varied[4]. Heavy metal toxicity has been shown to affect almost every organ system of the body with continued exposure[5]. Management of liver diseases is still a challenge to modern medicine. Modern medicine has little to offer for alleviation of hepatic ailments.

Most of the hepato-protective agents now available are expensive and hence a genuine need is felt to devise some cost effective drugs based on plant principles in this regard[6]. Numerous medicinal plants and various formulations are used for liver disorders in ethno medical practices as well as in traditional systems of medicine in India[7]. Many plants possess hepato-protective activity against carbon tetrachloride, ethanol, paracetamol, anti tubercular drugs and galactosamine induced liver damage in albino rats and hence a similar study mentioned is presented in this study.

*Cayratia carnosa* (Wall. Ex Wight) Gagnep. (Vitaceae) is a small fleshy tendril climber; tendrils short, slender and branched; leaves trifoliate, usually pubescent, leaflets dentate; flower buds globose; fruits fleshy berries; seeds pyriform or triangular, rounded and rugose on the back. The plant grows wildly throughout the Western Ghats, in India. The whole plant is used for tumors, fever, neuralgia and splenomegaly. It purifies the blood and is given for liver diseases, cardiac disorders, ulcers, wounds, dropsy and hemorrhoids. During the present study, an herbal drug *Cayratia carnosa* is chosen for the evaluation of hepato-protective activity which could not be attempted by any researcher earlier.

**MATERIALS AND METHODS**

**Plant materials**

The whole plant of *Cayratia carnosa* was collected for the study in the month of August, the flowering season period, from the wild source in the plains area of Ulloor of Thiruvananthapuram city, Kerala. *Cayratia carnosa* was authenticated by Dr. P. Brindha, Botanist, Department of Botany, Captain Srinivasa Murti Drug Research Institute for Ayurveda, Arumbakkam, Chennai, Tamilnadu, India.

**Preparation and selection of the extracts**

The whole plant of *Cayratia carnosa* after collection was washed in running tap water to remove the soil and adhering materials, shade dried, coarsely powdered and packed in the ssxshet extractor. The packed material was extracted successively with the following solvents: Petroleum ether, benzene, chloroform, acetone, ethanol and water. Total aqueous extract was also prepared separately by cold maceration method[8]. These extracts were filtered, distilled and concentrated. The concentrated extracts were dried under vacuum in desiccators containing anhydrous calcium chloride. Since the ethanol, aqueous and total aqueous extracts contains the maximum number of phytoconstituents, these extracts alone were selected for the pharmacological activity.

**Animals**

Male albino rats of Wistar strain was used for hepato-protective activity. These animals were taken from the inbred colony maintained in the animal house of our Institution. Male albino rats weighing 220 to 250g were used for the study. These animals were fed with pelleted diet manufactured by Amrut laboratory animal feed company, Sangli, Maharashtra and drinking water ad libitum. They were kept in 12hrs/12hrs light/ dark cycle for aclimatization towards the laboratory conditions before starting the experimental procedure. Ethical clearances for performing the experiments on animals were obtained from SB College of Pharmacy, Institutional Animal Ethical Committee with reference no: IAEC/SB/2004-2005/E7 (I) /252a dated 23/06/2004.

**Paracetamol Induced Hepato Toxicity in Albino Rats**

Hepato-protective activity was evaluated against paracetamol induced hepatic damage. Paracetamol toxicity is due to the formation of toxic metabolites (N-acetyl-p-benzoquinoneminine) through the action of cytochrome P450. Induction of cytochrome P450 – depletion of hepatic glutathione is a prerequisite for paracetamol induced hepatotoxicity[9]. Liver tissue, rich in both transaminases (SGOT and SGPT) contains more SGPT and SGOT while both transaminases are elevated in sera of patients with acute hepatic
Hepato-Protective Activity of Cayratia carnosa

Male albino rats of Wistar strain were selected and divided into nine groups of six animals each. They should be treated for seven days as follows:

Group I = Normal control (Normal saline 10 ml/kg)
Group II = Paracetamol 2.5 kg/kg.
Group III = Silymarin (25 mg/kg).
Group IV = Ethanolic extract 200 mg/kg
Group V = Aqueous extract 200 mg/kg
Group VI = Total Aqueous extract 200 mg/kg
Group VII = Ethanolic extract 400 mg/kg
Group VIII = Aqueous extract 400 mg/kg
Group IX = Total Aqueous extract 400 mg/kg

On the seventh day paracetamol suspension (2.5 g/kg) in 0.5% CMC was given by oral route to all the animals except animals in Group I. This dose is known to cause liver damage in albino rats[16]. After 36 hours, all the animals were sacrificed under ether anesthesia; blood was collected by cardiac puncture in sterile centrifuge tubes and allowed to clot. Serum was separated by centrifuging at 2500 rpm for 10 min and used for the estimation of SGOT, SGPT[17], alkaline phosphatase[18] and serum bilirubin[19] levels.

**Histopathological examination**

After the animals were sacrificed, the abdomen was cut open and the liver was removed. The ratio of wet liver weight per 100 g of body weight was calculated and recorded[20]. The liver excised was washed with normal saline. Initially the materials were fixed in 10% buffered neutral formalin and then with bovine solution. They were processed for paraffin embedding following the microtome technique[21]. The sections were processed in alcohol, xylene series and were stained with alun haematoxylin and eosin. The stained sections were examined microscopically for histopathological changes.

**Statistical Analysis**

All the values are expressed as Mean ± SEM. The data obtained from hepatoprotective study was subjected to one-way ANOVA followed by Dunnet’s test for statistical significance. P<0.05 is considered to be statistically significant.

**RESULTS AND DISCUSSION**

Effect of *Cayratia carnosa* on paracetamol induced hepatotoxicity in albino rats

The results of the hepatoprotective activity are presented in Table 1. Paracetamol causes significant increase in the activity of serum transaminases, alkaline phosphatase and bilirubin levels. Results indicate that various test extracts of *Cayratia carnosa* provides significant protection against the paracetamol induced toxic effect on liver. In paracetamol induced toxic hepatitis, toxicity begins with changes in endoplasmic reticulum, which results in the loss of metabolic enzymes located in intra cellular structure[22]. The blood samples of the paracetamol alone treated animals showed drastic increase in the levels of liver weight (5.05 g), SGOT (63.5 units/ml), SGPT (47 units/ml), alkaline phosphatase (29 units/ml) and total bilirubin (1.5 mg/dl) as compared to control group showing liver weight (2.7 g), SGOT (25 units/ml), SGPT (21 units/ml), alkaline phosphatase (7.5 units/ml) and total bilirubin (0.35 mg/dl). Elevation of serum enzyme and bilirubin levels is considered as an index of liver damage[23,24,25] and liver necrosis. Histopathological sections of the paracetamol treated animals showed focal necrosis and portal inflammation as compared to the intact arrangement of cells of the control animal.

Administration of silymarin and various extracts of *Cayratia carnosa* showed hepatoprotective effect against the toxic effects of paracetamol as shown in the Table 1. Among these extracts significant hepatoprotective was noticed in the group 7 animals treated with ethanolic extract at 400 mg/kg which was comparable to that of standard drug silymarin while the aqueous extract 200 mg/kg treated animals exhibited the least hepatoprotective activity. Histopathological profile of all the test extracts treated animals showed recovery against the paracetamol induced necrosis in their normal compact arrangement of hepatic cells as compared to control.

<table>
<thead>
<tr>
<th>Gr. No</th>
<th>Drug Treatment</th>
<th>Dose (mg/kg)</th>
<th>Liver weight g/100g body wt</th>
<th>SGOT units/ml (8-20)</th>
<th>SGPT units/ml (5-40)</th>
<th>Alkaline Phosphatase KA Units/ml (5-13)</th>
<th>Total Bilirubin mg/dl (0.2-0.55)</th>
<th>Direct Bilirubin mg/dl</th>
<th>Indirect Bilirubin mg/dl</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control (Saline)</td>
<td>10 ml/kg</td>
<td>2.70±0.062</td>
<td>25±1.29</td>
<td>21±1.29</td>
<td>7.5±0.866</td>
<td>0.35±0.065</td>
<td>0.125±0.017</td>
<td>0.225±0.039</td>
</tr>
<tr>
<td>2</td>
<td>Paracetamol</td>
<td>2.5 g/kg</td>
<td>5.05±0.124</td>
<td>63.5±1.707</td>
<td>47±1.29</td>
<td>29±1.291</td>
<td>1.5±0.129</td>
<td>0.095±0.026</td>
<td>1.405±0.123</td>
</tr>
<tr>
<td>3</td>
<td>Silymarin</td>
<td>25</td>
<td>3.15±0.053</td>
<td>25.5±1.5</td>
<td>21.5±0.957</td>
<td>65±0.5</td>
<td>0.375±0.063</td>
<td>0.1±0.085</td>
<td>0.275±0.058</td>
</tr>
<tr>
<td>4</td>
<td>Ethanolic extract</td>
<td>200</td>
<td>4.01±0.095</td>
<td>31±1.290</td>
<td>25±1.29</td>
<td>9.25±0.479</td>
<td>0.65±0.029</td>
<td>0.095±0.033</td>
<td>0.555±0.099</td>
</tr>
<tr>
<td>5</td>
<td>Aqueous extract</td>
<td>200</td>
<td>4.63±0.007</td>
<td>33.5±0.957</td>
<td>26±0.816</td>
<td>10.25±0.479</td>
<td>0.8±0.408</td>
<td>0.15±0.014</td>
<td>0.65±0.047</td>
</tr>
<tr>
<td>6</td>
<td>Total aqueous extract</td>
<td>200</td>
<td>4.35±0.045</td>
<td>32±0.816</td>
<td>25.5±0.957</td>
<td>9.5±0.289</td>
<td>0.7±0.408</td>
<td>0.085±0.018</td>
<td>0.615±0.071</td>
</tr>
<tr>
<td>7</td>
<td>Ethanolic extract</td>
<td>400</td>
<td>3.51±0.030</td>
<td>27±1.290</td>
<td>22±0.816</td>
<td>7.25±0.479</td>
<td>0.45±0.064</td>
<td>0.11±0.013</td>
<td>0.34±0.031</td>
</tr>
<tr>
<td>8</td>
<td>Aqueous extract</td>
<td>400</td>
<td>3.84±0.048</td>
<td>13±1.826</td>
<td>24±0.816</td>
<td>8.5±0.866</td>
<td>0.6±0.041</td>
<td>0.12±0.041</td>
<td>0.475±0.059</td>
</tr>
<tr>
<td>9</td>
<td>Total aqueous extract</td>
<td>400</td>
<td>4.05±0.157</td>
<td>29±1.290</td>
<td>23±1.291</td>
<td>8±0.408</td>
<td>0.55±0.065</td>
<td>0.125±0.018</td>
<td>0.425±0.057</td>
</tr>
</tbody>
</table>
i) \( P<0.001, \ P<0.01, \ P<0.02, \ P<0.05, \text{ n.s – non significant} \) Vs Control

ii) Values are Mean ± S.E.M

iii) One way ANOVA followed by Dunnet’s t-test.

iv) \( n=6 \) (Number of animals in each group)

v) Values in the parenthesis indicate the normal range of biochemical parameters.
CONCLUSION
The results obtained in this study provide a scientific support to the traditional folkloric use of Copratia cariosa as a hepato-protective agent. Further, isolation of active principles will be advantageous to produce novel bioactive constituents from these extracts which may possess more significance in the treatment of liver diseases and to elucidate its exact mechanism of action. Attempts are being made to isolate and characterize the active principle to which the hepatoprotective activity can be attributed.

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REFERENCES