EVALUATION OF HEPATOPROTECTIVE ACTIVITY OF TECTONA GRANDIS LINN.

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ARTICLE INFO:

Article history:
Received: 18 December 2013
Received in revised form: 15 March 2014
Accepted: 28 March 2014
Available online: 30 June 2014

Keywords:
Hepatic damage
Tectona grandis
Hepatoprotective
Hydroalcoholic
Biochemical parameters

ABSTRACT

Hepatic damage is associated with distortion of the metabolic functions of liver. Thus liver diseases remain as the serious health problems. The present study was aimed to evaluate the hepatoprotective activity of hydroalcoholic extract of Tectona grandis leaf. The phytochemical screening showed the presence of alkaloids, amino acids, carbohydrates, flavonoids, saponins, steroids, triterpenoids, and tannins. Evaluation of hepatoprotective activity of hydroalcoholic extract of Tectona grandis leaf was carried out at two dose levels 300 mg/Kg and 600 mg/Kg. Carbon tetrachloride induced hepatotoxic model was followed for investigation of hepatoprotective activity of Tectona grandis in wistar rats. The biochemical parameters (SGOT, SGPT and ALP) in rat serum were found to be increased in CCl4 induced hepatotoxicity model. Hydroalcoholic extract of Tectona grandis leaf was found to decrease the biochemical parameters (SGOT, SGPT and ALP) of rat serum at the doses of 300 mg/Kg and 600 mg/Kg. Hydroalcoholic extract of Tectona grandis leaf significantly (p≤0.05) and dose dependently produced hepatoprotective activity at the doses of 300 mg/Kg and 600 mg/Kg.

1. Introduction

Liver is a key organ regulating homeostasis within the body by various functions. Liver injury caused by toxic chemicals and certain drugs has been recognized as a toxicological problem. Hepatotoxicity is one of very common ailment resulting into serious debilities ranging from severe metabolic disorders to even mortality[1]. Liver diseases remain one of the major threats to public health and are a worldwide problem[2].

Medicinal plants play a key role in human health care. Scientific studies available on medicinal plants indicate that promising phytochemicals can be developed for many health problems[3]. Liver protective plants contain a variety of chemical constituents like phenols, coumarins, monoterpenes, glycosides, alkaloids and xanthenes[4]. Tectona grandis is a useful plant which has been credited with therapeutic properties to treat several diseases. Tectona grandis commonly known as Teak in English and Sagaun in hindi. Traditionally Tectona grandis is found to poses Sedative, Anthelmintic, anti-inflammatory, laxative, diuretic, emollient, demulcent properties[5-9]. The various plant parts are found to contain photochemicals like Quinones (Tectoquinone, lapachol, deoxylapachol and its isomer, tectoleafoquinone, anthraquinone–napthaquinone pigment). Steroidal compounds (Squalene, polyisoprene-α-tolyl methyl ether, betulinic acid, tectogradone, monoterpene, Apocarotenoids: tectoionols-A, tectoionols-B), Glycosides (Anthraquinone glycosides), Phenolic acids(Tannic acid, gallic acid, ferulic acid, caffeic acid and ellagic acid), Flavonoids (Rutin and quercitin). Tectona grandis Linn leaf is also reported to contain carbohydrates, alkaloids, tannins, sterols, saponins, proteins, calcium, phosphorus, crude fiber and also to contain dye (yellowish-brown or reddish)[10-13]. The objective of present study was to evaluate the hepatoprotective activity of hydroalcoholic extract of Tectona grandis leaf against carbon tetra chloride induced hepatotoxicity in rats.

2. Materials and methods

2.1 Plant Material

Fresh leaves of the plant Tectona grandis (Linn.) were obtained from herbal garden at National Botanical Researh Institute, Lucknow. The plant material was identified and internal authentication done by Dr. Vijay Kumar, Scientist E-II, Ethnopharmacology and Pharmacognosy division. The specimen was deposited in the departmental herbarium of National Botanical Research Institute, Lucknow, India for future reference. The collected leaves were dried in shade in tray drier at 25°C, crushed in grinder to coarse powder and stored in a well closed container for further studies.
2.2 Preparation of hydroalcoholic extract of Tectona grandis leaf

The leaves of Tectona grandis were collected, dried in the shade, powdered, and weighed (1 kg). The powdered plant material was exhaustively extracted with hydroalcoholic solvents (methanol: 50% v/v and water: 50% v/v) by cold maceration for 3 days at room temperature. The extract was separated by filtration and concentrated on rotavapour (Buchi, USA) and then dried in freeze/dry system 4.5 at high vacuum and at temperature -40°C (yield 5.4% w/w). The extract was subjected to phytochemical and pharmacological investigations.

2.3 Preliminary phytochemical screening

The hydroalcoholic extract of Tectona grandis leaf was subjected to preliminary phytochemical screening[14].

2.4 Animals

Male Wistar rats, weighing 200-250 gm were procured from the central animal house of Central Drug Research Institute Lucknow, India. The animals were kept in the departmental animal house in polyacrylic cages with not more than six animals per cage and maintained under standard laboratory conditions at 26 ± 2 ºC and 44 – 56% relative humidity, light and dark cycle of 10 and 14 hrs respectively for 1 week before and during the experiment for accclimatization. The animals were provided with standard rodent pellet diet (Amrut, India) and the food was withdrawn 18-24 hours before the experiment though water was allowed ad libitum. All experiments were performed in the morning according to current guidelines for the care of laboratory animals and the ethical guidelines for investigations of experimental pain in conscious animals[15].

2.5 Animal models for Hepatoprotective activity

2.5.1 Carbon Tetra chloride (CCl4) Induced Hepatotoxicity

The animals were divided into five groups of six Wistar albino rats each. The animals were fasted for 24 h prior to carbon tetra chloride treatment. Control Group I received normal saline (0.9% sodium chloride solution) 5 ml/Kg orally. All the animals of group II to V received carbon tetra chloride diluted with olive oil (1:1) at dose of 1 ml/Kg, subcutaneously for two successive days (1st and 3rd day), Group II animals were maintained as negative control group without any drug treatment. Group III animals were treated with Silymarin (100 mg/Kg, orally) which served as standard group. Group IV and V animals were treated with 300 mg/Kg and 600 mg/Kg hydro alcoholic extract of Tectona grandis leaf respectively by oral route. The vehicle or drug treatment was carried out orally from 1st day to 5th day with concurrent administration of carbon tetra chloride on 2nd and 3rd day. During the period of drug treatment the rats were maintained under normal diet and water ad libitum. The animals of all the groups were sacrificed by light ether anaesthesia on 6th day[16]. The blood sample of each animal was collected separately by carotid artery into sterilized dry centrifuge tubes and allowed to coagulate for 30 min. Serum was separated by centrifugation 3000 rpm for 15 min. The serum was used to estimate serum glutamate pyruvate transaminase (SGPT), serum glutamate oxaloacetate transaminase (SGOT)[17], serum alkaline phosphatase (ALP)[18]. Livers were removed and preserved in 10% formalin solution for histopathological studies.

2.5.2 Histopathological observation

Liver tissue collected were used for the preparation of histopathological slides by using microtome and were suitably stained and observed under microscope for architectural changes seen during CCl4 induced hepatotoxicity in hydroalcoholic extract of Tectona grandis leaf treated groups and control group.

2.5.3 Biochemical analysis

The collected blood samples were used for the analysis of biochemical markers serum glutamate pyruvate transaminase (SGPT), serum glutamate oxaloacetate transaminase (SGOT)[17], serum alkaline phosphatase (ALP)[18].

2.5.4 Statistical analysis

The mean ± S.E.M. was calculated for each parameter. Total variations, present in a set of data were estimated by one way analysis of variance (ANOVA), followed by Tukey’s test p≤ 0.05 was considered as statistically significant when compared to control group.

3. Results

The effect of hydroalcoholic extract of Tectona grandis leaf on weight of liver, weight of body and biochemical parameters in Carbon Tetra chloride intoxicated rats are shown in Table 1.

Table 1. Effect of hydroalcoholic extract of Tectona grandis leaf on weight of liver, weight of body and biochemical parameters in Carbon Tetra chloride intoxicated rats

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatments</th>
<th>Dose (Kg⁻¹)</th>
<th>Body Weight (gm)</th>
<th>Liver Weight (gm)</th>
<th>SGPT (U/L)</th>
<th>SGOT (U/L)</th>
<th>ALP (U/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control (Normal Saline)</td>
<td>5 ml</td>
<td>239.42±3.08</td>
<td>8.74±0.28</td>
<td>57.16±2.54</td>
<td>152.00±4.59</td>
<td>125.50±4.68</td>
</tr>
<tr>
<td>II</td>
<td>Negative control (CCl4)</td>
<td>1 ml</td>
<td>257.90±6.27a</td>
<td>10.58±0.15a</td>
<td>353.00±2.30a</td>
<td>466.17±7.38a</td>
<td>376.67±5.73a</td>
</tr>
<tr>
<td>III</td>
<td>Standard (Silymarin)</td>
<td>100mg</td>
<td>243.00±4.09b</td>
<td>8.97±0.62b</td>
<td>80.67±2.31b</td>
<td>223.67±4.96b</td>
<td>171.50±7.01b</td>
</tr>
<tr>
<td>IV</td>
<td>Test-I (Tectona grandis leaf)</td>
<td>300mg</td>
<td>254.85±9.05ab</td>
<td>9.73±0.19ab</td>
<td>161.67±4.45ab</td>
<td>294.18±6.62ab</td>
<td>272.83±6.79ab</td>
</tr>
<tr>
<td>V</td>
<td>Test-II (Tectona grandis leaf)</td>
<td>600mg</td>
<td>248.00±1.58 ab</td>
<td>9.15±1.03ab</td>
<td>115.00±4.86ab</td>
<td>252.65±6.28ab</td>
<td>229.00±5.17ab</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SEM of 6 rats in each group. p<0.001 when compared to control and p<0.001 when compared to CCl4 treated group (negative control)
4. Discussion

The CCl₄ has been used as a tool to induce hepatotoxicity in experimental animals. This toxic chemical caused peroxidative degradation in the adipose tissue resulting in fatty infiltration of the hepatocytes. The increase in the levels of serum bilirubin reflected the depth of jaundice and the increase in transaminases and alkaline phosphatase was the clear indication of cellular leakage and loss of functional integrity of the cell membrane. Administration of hydroalcoholic extract of *Tectona grandis* leaf showed significant hepatoprotective activity, which was comparable with the standard drug silymarin. Normally biochemical parameters like SGOT, SGPT and ALP are present in high concentration in liver. The reduced concentrations of SGOT, SGPT and ALP as a result of plant extract administration observed during the study might probably be due to the presence of flavonoids. Liver protective herbal drugs contain a variety of chemical constituents like phenols, coumarins, lignans, essential oil, monoterpenes, carotinoids, glycosides, flavanoids, organic acids, lipids, alkaloids and xanthenes.

5. Conclusion

The pharmacological studies provided the data of hydroalcoholic extract of *Tectona grandis* Linn. leaf which will help in identifying and differentiating it from other species. The presence of a variety of medicinally active chemical substances such as sterols, triterpenoids, saponins, alkaloids, flavonoids and tannins in *Tectona grandis* plant gives a lead for further studies to identify and to isolate particular components responsible for its medicinal activity. CCl₄ induced alteration on protein metabolism and hepatic antioxidant defense system were normalized by administration of hydroalcoholic extract of *Tectona grandis* leaf indicating its possible cytoprotective role against CCl₄ induced hepatotoxicity. The present study gives scientific support to the traditional use of the plant as hepatoprotective agent in folklore medicine. The hepatoprotective activity of hydroalcoholic extract of *Tectona grandis* leaf may be due to reduction of peroxidative degradation in the adipose tissue and SGPT, SGOT and ALP mediated oxidative stress in the hepatotoxic liver of wistar rat. However, further investigations are needed to evaluate the potential usefulness of this plant in clinical conditions associated with liver damage.

References


Source of support: Nil, Conflict of interest: None Declared