Though liver disease is a worldwide problem, available remedies are scarce. Conventional drugs used in the treatment of liver diseases are inadequate and can have serious adverse effects. Due to lack of reliable allopathic drug to cure liver damage researchers look for herbal drugs with better hepatoprotective action. Several studies have been carried out to see hepatoprotective effect of plants such as Semecarpus anacardium, Aerva lanata, Costus speciosus and Cleome viscosa.

Studies suggest that hepatoprotective effect of Tamarindus indica is due to the presence of flavonoids, ascorbic acid and -carotene. Hepatoprotective effect of Tamarindus indica (Family: Caesalpiniaceae), commonly known as tentul, has been studied using aqueous extracts of leaf, fruit and seed. In the present study ethanolic extracts of leaf and seed of Tamarindus indica were used to see whether the ethanolic extracts is more effective than aqueous extracts and its effect was compared with vitamin E.
Materials and Methods

Plant Materials

Leaves and seeds of Tamarindus indica were procured from Sirajganj and identified by plant taxonomy Unit of Bangladesh National Herbarium with accession no. DACB-35524, which was deposited to the Herbarium.

Preparation of Plant extracts

Leaves and seeds were shade dried and powdered by electric blender and grinder machine. The powders were soaked separately in 95% ethanol. The extract so obtained was concentrated in vacuum rotatory evaporator at 40-50°C until a paste was formed. Extract paste was freeze dried.

Drugs and Chemicals

Paracetamol powder was obtained from Kumudini Pharmaceuticals Ltd, Bangladesh. Propylene glycol was used as solvent for paracetamol powder. Vitamin E solution was collected from Drug international Ltd, Bangladesh.

Animals

A total of 30 male Long Evans rats (150-180 gm) were used for this study. The animals were kept in well ventilated room in the animal house of Dhaka Medical College. A 12 hr light/12 hr dark cycle was maintained. They were given standard food pellets and allowed drinking water ad libitum. Ethical clearance for the use of animals was obtained from the Committee constituted for the purpose.

Methodology

A total of thirty rats were taken and divided into five groups. Each group having six rats (n=6). Control group received normal diet ad libitum. Only paracetamol group received paracetamol (1500 mg/kg) for 7 days and three groups received paracetamol (1500 mg/kg) for first 7 days followed by ethanolic extract of leaf (1250 mg/kg) and seed (1250 mg/kg) of Tamarindus indica and vitamin E (500 mg/kg) for next 5 days.

After 24 hours of last treatment, all rats were anesthetized with light chloroform and blood was collected by cardiac puncture and serum was separated for estimations of serum alanine aminotransferase (ALT), serum aspartate aminotransferase (AST), serum alkaline phosphatase (ALP) and serum bilirubin. Liver was taken for histopathological examination.

Histopathological Study

Liver tissue was taken and fixed in 10% formalin and sections of liver tissue were embedded in paraffin and made blocks. Serial sections of blocks 3µ-5µ thickness were made and stained with haematoxylin and eosin and examined under microscope.

Statistical Analysis

The values were expressed as mean± SD. The statistical analysis was carried out by unpaired student's 't' test and p<0.05 was considered as significant.

Results

In rats pretreated with paracetamol, serum bilirubin, ALT, AST and ALP increased significantly as compared to control group (Table I). Significant decrease in serum bilirubin, ALT, AST and ALP was observed following administration of ethanolic extract of leaf and seed of Tamarindus indica and vitamin E (Table I). The rate of decline in serum enzyme level following administration of ethanolic extract of leaf and seed of T. indica was same as that of vitamin E. Histopathological examination of liver tissue in Paracetamol administered rats showed massive liver tissue necrosis with loss of cellular architecture and infiltration of neutrophil, macrophage and lymphocyte (Fig: 2). An almost return to normal architecture of hepatic tissue was observed in rats that received ethanolic extract of leaf and seed of Tamarindus indica (Fig: 3 & 4). Similar texture and cell arrangement were observed in the liver section of rats treated with vitamin E (Fig: 5).
Table 1: Serum bilirubin, ALT, AST and ALP level in Paracetamol pretreated (7 days) rats that received ethanolic extract of leaf and seed of Tamarindus Indica and vitamin E.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum bilirubin (mg/dl)</th>
<th>Serum ALT (U/L)</th>
<th>Serum AST (U/L)</th>
<th>Serum ALP (U/L)</th>
</tr>
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<tbody>
<tr>
<td>Control</td>
<td>0.45±0.13</td>
<td>41.33±4.45</td>
<td>36.33±3.44</td>
<td>90.00±12.59</td>
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<tr>
<td>Only Paracetamol</td>
<td>1.06±0.40***</td>
<td>193.00±12.87***</td>
<td>166.00±19.67***</td>
<td>428.00±33.66***</td>
</tr>
<tr>
<td>Leaf extract T. indica</td>
<td>0.95±0.17**</td>
<td>61.33±2.50 ***</td>
<td>50.33±1.83 ***</td>
<td>174.00±22.45 ***</td>
</tr>
<tr>
<td>Seed extract T. indica</td>
<td>0.57±0.20*</td>
<td>50.17±4.54 ***</td>
<td>53.00±5.59 ***</td>
<td>178.00±27.80 ***</td>
</tr>
<tr>
<td>Vitamin E</td>
<td>0.52±0.18*</td>
<td>55.33±8.08 ***</td>
<td>47.67±6.06 ***</td>
<td>142.00±17.66 ***</td>
</tr>
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</table>

n = 6. All the drugs were administered orally through Ryles tube. Data expressed as mean ± SD. **P<0.01. ***P<0.001.

Fig-1: Photomicrographs 40X showing the normal hepatic architecture in control group.

Fig-2: Photomicrographs 40X showing Paracetamol induced hepatic necrosis and small number of inflammatory cells.

Fig-3: Photomicrographs 40X showing normal hepatic architecture following administration of leaf extract of Tamarindus indica in Paracetamol induced hepatotoxicity.

Fig-4: Photomicrographs 40X showing normal hepatic architecture following administration of seed extract of Tamarindus indica in Paracetamol induced hepatotoxicity.

Fig-5: Photomicrographs (Magnification at 10X and 40X objectives) showing normal hepatic architecture following administration of Vitamin E in Paracetamol induced hepatotoxicity.
Discussion

Significant increase in serum bilirubin, ALT, AST and ALP following administration of paracetamol is an indicator of hepatotoxicity. Assessment of liver damage can be assessed by estimation of serum ALT, AST and ALP. Necrosis results in the release of these enzymes into circulation, therefore, it can be measured in serum. High levels of AST indicate liver damage, ALT catalyses the conversion of alanine to pyruvate and glutamate and is released in similar manners, therefore, ALT is more specific to liver and is thus a better parameter for detecting liver damage.

The results in the present study showed a significant damage to liver tissue following administration of paracetamol, confirmed by histopathological examination of liver tissue that showed massive necrosis and infiltration of macrophages and lymphocytes.

Paracetamol causes acute liver damage due to depletion of glutathione and formation of highly reactive metabolite N-acetyl Parabrenzoquinone-imine (NAPQI). NAPQI arylates essential nucleophilic macromolecules within hepatocytes, forming stable acetaminophen-protein adducts which are responsible for Acetaminophen induced hepatotoxicity. Elevated enzyme levels showed loss of functional integrity of hepatocytes. Administration of ethanolic extracts of leaf and seed of Tamarindus indica decreased liver enzymes level and helped return of liver tissue to almost normal architecture on histopathological examination, which is same as that of vitamin E. Similar other studies have been carried out using aqueous extract of leaf, seed and fruit extracts of Tamarindus indica and similar results were observed. From this study it can be assumed that both Tamarindus indica and vitamin E are hepatoprotective and their hepatoprotective effect is almost equal. Hepatoprotective effect of vitamin E is due to its antioxidant property. Hepatoprotective effect of Tamarindus Indica has been attributed to the presence of flavonoids, polyphenol, -carotene and ascorbic acid. A number of scientific reports indicate that flavonoids, -carotene, ascorbic acid have protective effect on liver due to their antioxidant properties.

It seems that Tamarindus indica, a very commonly used food, can be applied for treatment of drugs or chemical induced hepatotoxicity. Use of natural products cause less adverse effect compared to synthetic analogues. Tamarindus indica is easily available, cheap and its fruit is consumed as a popular food. Considering its availability and cheapness and its comparable effectiveness to vitamin E, it might be considered as a hepatoprotective agent and applied in the treatment of drug related hepatotoxicity. Before clinical application further studies on its beneficial hepatoprotective effect should be carried out.

References

Biochemical Factors associated with Breast Cancer


