Antihepatotoxic and antioxidant potential of *Hedyotis corymbosa* extract against antitubercular drugs induced hepatotoxicity in experimental animal

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**Abbreviations:**  
*Hedyotis corymbosa*: HCE, Total bilirubin: TBL, Albumin: ALB; Total protein: TP, Lactate dehydrogenase: LDH, Cholesterol: CHL

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**Abstract**  
To assess the antioxidant and hepatoprotective effect of *Hedyotis corymbosa* extract against antitubercular drug-induced liver toxicity in experimental animals. Methanolic extract of *Hedyotis corymbosa* (HCE, 100 and 200 mg/kg body weight) was administered daily for 35 days. Liver toxicity was induced by combination of three antitubercular drugs [Isoniazid 7.5 mg/kg, Rifampicin 10 mg/kg and Pyrazinamide 35 mg/kg] given orally as suspension for 35 days in rats. The hepatoprotective activity was assessed using various biochemical parameters like aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), total bilirubin (TBL), albumin (ALB), total protein (TP), lactate dehydrogenase (LDH), and serum cholesterol (CHL). Meanwhile, in vivo antioxidant activities as lipid peroxidation (LPO), reduced glutathione (GSH), superoxide dismutase (SOD) and catalase (CAT) were measured in rat liver homogenate along with histopathological examination. Obtained results demonstrated that the treatment with HCE significantly and dose-dependently prevented drug induced increase in serum levels of hepatic enzymes. Furthermore, HCE significantly reduced...
the lipid peroxidation in the liver tissue and restored activities of defence antioxidant enzymes towards normal levels. Histopathology of the liver tissue showed that HCE attenuated the hepatocellular necrosis and led to reduction in inflammatory cells infiltration. The results of this study strongly indicate the hepatoprotective effect of HCE against antitubercular drugs and there by scientifically support its traditional use.

**Citation:**

### 1. Introduction

Liver diseases have become a global problem and about 20,000 deaths occur every year due to liver disorders and it is well known that free radicals cause cell damage through mechanisms of covalent binding and lipid peroxidation with subsequent tissue injury. Antioxidants are vital substances which possess the ability to protect the body from damage caused by free radical induced oxidative stress (Chaudhary et al., 2012; Shakya et al., 2012;). The pathogenesis of hepatic diseases as well as the role of oxidative stress and inflammation therein is well established (Tacke et al., 2009) and accordingly blocking or retarding the chain reactions of oxidation and inflammation process could be a promising therapeutic strategy for prevention and treatment of liver injury. Detoxification reactions (phase I and phase II) metabolize xenobiotics with the aim of increasing substrate hydrophilicity for excretion. Drug-metabolizing enzymes detoxify many xenobiotics but bioactivate or increase the toxicity of others. In the case of bioactivation, liver is the first organ to be exposed to the damaging effects of the newly formed toxic substance. Therefore protective mechanisms relevant to the liver are of particular interest. Effectively, herbal products are widely used in the treatment of hepatic disorders all over the world (Amat et al., 2010).

*Hedyotis corymbosa* (L.) Lam. Syn. *Oldenlandia corymbosa* (L.) Lam. (Rubiaceae) is a weedy herb, widely distributed throughout India. It is commonly known as ‘Parppatakappul’ in traditional medicine of Kerala. There are approximately 180 species recorded of which 35 were identified in Malaysia (Ang et al., 2010). *Hedyotis corymbosa* is extensively used in modern Chinese practice for the treatment of viral infections, cancer, syndromes involving “toxic heat”, acne, boils, skin ailments, appendicitis, hepatitis, eye diseases and bleeding (Chen et al., 1994). They call it ‘Peh-Hue-Juwa-Chi-Cao’ (Valarmathi et al., 2011). The plant is used for treating venomous bites. It is bitter, acrid, cooling, febrifugal, pectoral, anthelmintic, diuretic, depurative, diaphoretic, expectorant, digestive and has stomachic properties (Rathi et al., 2009). It is given in jaundice, and other diseases of the liver and heat eruptions, vitiated conditions of pitta, hyperdyspia, giddiness, dyspepsia, flatulence, colic, constipation, helminthiasis, leprosy, skin diseases, cough, bronchitis, necrosis, nervous depression caused by deranged bile and hepatopathy.

Three new iridoid glycosides (Sasikumar et al., 2010), nine iridoid and lignin glycosides and rutin have been isolated from the whole plant (Shibano et al., 2008). To the best of our knowledge there was lack of scientific reports available in support of its traditional claim of hepatoprotective potential. So far there has been only two researches reported on hepatoprotective effect against carbon tetrachloride (Wang et al., 2011) and paracetamol (Sultana et al., 2010) induced liver damage in rats has been investigated. Therefore the present study was designed to demonstrate the effect of *Hedyotis corymbosa* extract (HCE) against the combination of three anti-tubercular drugs [isoniazid (I) -7.5 mg/kg, Rifampicin (R) -10 mg/kg and Pyrazinamide (P) -35 mg/kg] induced hepatic damage in experimental animals.

### 2. Objective of Research

Objective of this research is to assess the antioxidant and hepatoprotective effect of *Hedyotis corymbosa* extract against antitubercular drug-induced liver toxicity in experimental animals.

### 3. Materials and Methods

Drug-induced liver injury is a major health problem that challenges not only health care professionals but also the pharmaceutical industry and drug regulatory agencies. There is a growing interest in herbal remedies because of their effectiveness, minimal side effects in clinical experience and relatively low cost. Therefore, present study was designed...
to demonstrate the anti-hepatotoxic and antioxidant potential of *Hedyotis corymbosa* against anti-tubercular drugs induced acute hepatopathy in experimental animals.

### 3.1 Chemicals
All the chemicals were used analytical grade and procured from Sigma chemicals Co., USA and Qualigens fine chemicals, Mumbai, India.

### 3.2 Preparation of plant extract
The whole plants of *Hedyotis corymbosa* were collected from campus garden of National Botanical Research Institute, Lucknow, India in December 2010. The plant material was identified and authenticated and the voucher specimen was deposited in the institutional herbarium. The whole plants of *Hedyotis corymbosa* were washed thoroughly in tap water, shade dried and powdered. The powder (100 g) was successively extracted with 1000 ml of methanol overnight with constant stirring. The filtrate was then concentrated and the solvent was evaporated under reduced pressure in a rotary evaporator. The yield of the extract was found to be 0.42% (w/w). This crude extract was referred to as HCE. The extract obtained was further subjected to pharmacological investigation.

### 3.3 Animals
Wistar rats weighing (150-170 g) and Swiss albino mice (25-30 g) of either sex were procured from CDRI, Lucknow. They were kept in departmental animal house in well cross ventilated room at 22 ± 2° C with light and dark cycles of 12 h for 1 week before and during the experiments. Animals were provided with standard rodent pellet diet (Amrut, India) and the food was withdrawn 18-24 h before the experiment though water was given *ad libitum*. All studies were performed in accordance with the guide for the care and use of laboratory animals, as adopted and promulgated by the Institutional Animal Care Committee, CPCSEA, India.

### 3.4 Behavioural and toxicological effects
Two groups of 10 mice were treated with graded doses of the HCE (100 and 200 mg/kg, p. o.). One group was maintained as control and was given 0.5% Tween-80. They were observed continuously for 1 h for any gross behavioural changes and death, if any and then, intermittently for the next 6 h, and then again at 24 h after dosing with HCE (Sultana et al., 2010).

### 3.5 Antitubercular drugs induced Hepatotoxicity
The animals were divided into five groups, each group had six animals. Group I (control) animals were administered a single daily dose of carboxymethyl cellulose (1 ml of 1%, w/v, p.o. body weight). Group II rats were administered a combination of three anti-tubercular drugs [isoniazid (I) -7.5 mg/kg, Rifampicin (R) -10mg/kg and Pyrazinamide (P) -35 mg/kg] for 35 days by intra-gastric administration (Hussain et al., 2012) in sterile saline which severed as disease control while group III and IV received orally 100 and 200 mg/kg body weight of HCE in (1%, w/v, CMC) respectively, 45 min prior to antitubercular drugs challenge for 35 days. Group V received Silymarin the known hepatoprotective compound (Sigma Chemicals Company, USA) at a dose of 100 mg/kg, p.o., daily for 35 days, 45 min prior to antitubercular drugs challenge as a reference (Padma et al., 1998). On completion of the experimental period the blood was collected, the animals were sacrificed and liver samples were collected.

### 3.6 Assessment of hepatoprotective activity
The collected blood was allowed to clot and serum was separated at 2500 rpm for 15 min and the biochemical parameters like serum enzymes: aspartate aminotransferase (AST, U/L), alanine aminotransferase (ALT, U/L) (Reitman et al., 1957), alkaline phosphatase (ALP, U/L) (King et al., 2009), total bilirubin (mg/dL) (Malloy et al., 1937), albumin (ALB, gm/dl) and total protein (TP, gm/dl) (Lorry et al., 1951), lactate dehydrogenase (LDH, U/L) (Kornberg et al., 1995) and total cholesterol (CHL, gm/dl) (Varley et al., 1980).

### 3.7 Assessment of antioxidant parameters
#### 3.7.1 Assessment of lipid peroxidation (LPO)
The dissected out liver samples were washed immediately with ice cold saline to remove as much blood as possible. Liver homogenized (5%) in ice cold 0.9% NaCl with a Potter-Elvenhjem glass homogenizer. The homogenate was centrifuged at 800 rpm for 10 min and the supernatant was again centrifuged at 12,000 for 15 min and the obtained mitochondrial fraction was used for the estimation of LPO (Das et al., 1993). A volume of the homogenate (0.20 ml) was transferred to a vial and was mixed with 0.2 ml of a 8.1% (w/v) sodium dodecyl sulphate solution, 1.5 ml of a 20% acetic acid solution (adjusted to pH 3.5 with NaOH) and 1.5 ml of a 0.8% (w/v) solution of thiobarbituric acid (TBA) and the final volume was adjusted to 4 ml with distilled water.
Each vial was tightly capped and heated in a boiling water bath for 60 min. The vials were then cooled under running water. Equal volumes of tissue blank or test samples and 10% trichloroacetic acid were transferred into a centrifuge tube and centrifuged at 1000 rpm for 10 min. The absorbance of the supernatant fraction was measured at 532 nm (Beckman DU 650 spectrometer).

Control experiment was processed using the same experimental procedure except the TBA solution was replaced with distilled water (Jamal et al., 1985). Malondialdehyde (MDA) is an end product of lipid peroxidation, which reacts with thiobarbituric acid to form pink chromogen thiobarbituric acid reactive substance. 1, 1, 3, 3-tetra ethoxypropan was used as standard for calibration of the curve and is expressed as mmole/mg protein.

3.7.2 Assessment of catalase and superoxide

The liver tissue was homogenized (5%) and mitochondrial fraction was prepared as described above. Decomposition of H2O2 in presence of catalase (CAT) was followed at 240 nm (Aebi et al., 1984). One unit (U) of catalase was defined as the amount of enzyme required to decompose 1μmol of H2O2 per min, at 25° C and pH 7.0. Results are expressed as units (U) of CAT activity/mg protein. Superoxide dismutase (SOD) activity was estimated by the inhibition of nicotinamide adenine dinucleotide reduced)-phenazine methosulphate–nitrobluetetrazolium reaction system as described by Nishikimi et al. (1972) and as adapted by Kaekkar et al. (1972). One unit of the enzyme is equivalent to 50% inhibition in the formazan formation in 1 min at room temperature (25 ± 2° C) and the results have been expressed as units (U) of SOD activity/mg protein.

3.7.3 Assessment of reduced glutathione (GSH) activity

The concentration of GSH was determined by the method of Anderson (1985) based on the development of a yellow colour when 5, 5-dithiobis (2-nitrobenzoic acid) is added to compounds containing sulfhydryl groups. The reaction mixture contained equal volumes of 4% sulfosalicylic acid and tissue samples homogenized in 4 vol. of ice cold 0.1 m/l phosphate buffer (pH 7.4). The method used for estimating GSH in this study also measures non-protein sulfhydryl concentration inclusive of GSH. However, 80–90% of the non-protein sulfhydryl content of the cell represents free endogenous GSH. Enzyme activity was expressed as milligram per hundred gram (Amresh et al., 2007).

3.8 Histopathological studies

For histological studies, the liver tissues were fixed with 10% phosphate buffered neutral formalin, dehydrated in graded (50-100%) alcohol and embedded in paraffin. Thin sections (5 μM) were cut and stained with routine hematoxylin and eosin stain for photomicroscopic assessment. The initial examination was qualitative, with the purpose of determining histopathological lesions in liver tissue.

3.9 Statistical analysis

The values were represented as mean ± S.E.M. for six rats. Analysis of variance (ANOVA) test was followed by individual comparison by Newman-Keuls test using Prism Pad software (Version 5.0) for the determination of level of significance. The values of p<0.05 was considered statistically significant.

4. Results and Discussion

4.1 Behavioural and toxicological effects

_Hedyotis corymbosa_ produces no behavioural and toxicological effects at 100 and 200 mg/kg. Therefore 200 mg/kg dose of extract were selected as therapeutic highest dose and just half of its as lowest dose (100 mg/kg) in this study.

4.2 Effect of HCE on serum AST, ALT, ALP, TB, ALB, TP, LDH and cholesterol level

The effect various doses of HCE were studied on serum marker enzymes and total bilirubin in antitubercular drugs intoxicated animals. Hepatic injury induced by antitubercular drugs caused significant changed in marker enzyme as AST by 293.31%, ALT by 381.29%, ALP by 153.20%, total bilirubin by 365.21%, albumin by 34.93%, total protein by 99.41%, LDH by 69.57% and CHL by 101.33% compared to control group. The percentage protection in marker enzyme of treated groups at 100 mg/kg was 165.17%, 113.19%, 57.54%, 45.14%, 17.54% and 101.33% respectively compared to toxic group.

For histological studies, the liver tissues were fixed with 10% phosphate buffered neutral formalin, dehydrated in graded (50-100%) alcohol and embedded in paraffin. Thin sections (5 μM) were cut and stained with routine hematoxylin and eosin stain for photomicroscopic assessment. The initial examination was qualitative, with the purpose of determining histopathological lesions in liver tissue.
5.78 (P<0.01), 37.62 (P<0.001), TP 38.71 (P<0.001), 59.48 (P<0.001), LDH 14.55 (P<0.001), 23.15 (P<0.001) and CHL 33.27 (P<0.001), 44.84 (P<0.001) which is almost comparable to the group treated with silymarin, a potent hepatoprotective drug used as reference standard (Table 1).

4.3 Estimation of LPO, GSH, SOD and CAT
The results in Table 2 showed clear significant percentage change in the antioxidant levels of LPO in antitubercular drugs intoxicated rats as 290.90 (P<0.001) compared to control group. Treatment with HCE at the doses of 100 and 200 mg/kg significantly prevented this heave in levels and the percentage protection in LPO were 21.7 (P<0.05) and 58.91 (P<0.001) respectively. The GSH, SOD and CAT content had significantly increased in HCE treated groups whereas antitubercular drugs intoxicated group had shown significant decrease in these parameters compared to control group.

The percentage changed of GSH, SOD and CAT in antitubercular drugs intoxicated group were as 54.65 (P<0.001), 65.22 (P<0.001) and 40.27 (P<0.001) respectively. The percentage protection in GSH as 43.58 (P<0.05), 82.05 (P<0.001) and SOD 14.99 (n), 131.91 (P<0.01) while in CAT 26.11 (P<0.05), 45.92 (P<0.01) at the doses levels 100 and 200 mg/kg, respectively. In different doses level of HCE 200 mg/kg has shown maximum protection which was almost comparable to those of the normal control and silymarin.

4.4 Histopathological observations
The histological observations basically support the results obtained from serum enzyme assays. Liver section of normal control rat shows central vein surrounded by hepatic cord of cells while in anti-tubercular drugs treated rats showing massive fatty changes, focal necrosis with portal inflammation, loss of cellular boundaries and ballooning formation.

Whereas liver section of rats treated with 100 mg/kg of HCE showing mild focal necrosis but massive fatty changes with mild central vein congestion and less inflammatory cells and less ballooning. Liver section of rats treated with 200 mg/kg of HCE showing less inflammatory cells, absence of necrosis and congestion in central vein and showing regeneration of hepatocytes around central vein toward normal liver architecture possessing higher hepatoprotective action (Figure. 1).

In the present investigation, *Hedyotis corymbosa* (HCE) was evaluated for the hepatoprotective activity using antitubercular drugs induced liver toxicity in rat. Drug-induced liver toxicity is a potentially serious adverse effect of the currently used antitubercular chemotherapeutic regimens containing isoniazid, rifampicin and pyrazinamide. All these drugs are potentially hepatotoxic independently, when given in combination their toxic effects are enhanced in a synergistic manner. The conversion of monoacetyl hydrazine, a metabolite of isoniazid, to a toxic metabolite via cytochrome *P*<sub>450</sub> leads to hepatotoxicity. Rifampicin induces cytochrome *P*<sub>450</sub> enzyme causing an increased production of toxic metabolites from acetyl hydrazine (AcHz). Rifampicin can also increase the metabolism of Isoniazid to isonicotinic acid and hydrazine, both of which are hepatotoxic. The plasma half life of AcHz (metabolite of I) is shortened by Rifampicin and AcHz is quickly converted to its active metabolites by increasing the oxidative elimination rate of AcHz, which is related to the higher incidence of liver necrosis caused by I and R in combination (Hussain et al., 2003). Pyrazinamide (P), in combination with I and R is also, associated with an increased incidence of hepatotoxicity (Mitra et al., 1998).

In addition to these mechanisms; oxidative stress induced hepatic injury is one of the important mechanisms in hepatotoxicity produced by antitubercular drugs (Kale et al., 2003). The results of the present study suggested that HCE possess hepatoprotective activity against the hepatotoxicity induced by the combination of three antitubercular agents. The present study revealed a significant increase in the level of AST, ALT, ALP, serum bilirubin, LDH, CHL while decrease in ALB and total protein levels of group II on exposure to antitubercular drugs, indicating considerable hepatocellular injury. Elevated levels of these enzymes in serum are presumptive markers of drug induced necrotic lesions in the hepatocytes (Amr et al., 2005). Enhanced susceptibility of hepatocytes cell membrane to antitubercular drugs induced peroxidative damage might have resulted in increased release of these diagnostic marker enzyme levels into the systemic circulation (Santosh et al., 2000). The activity of ALT and AST are sensitive indicators of acute hepatic necrosis, and the ALP level is known to be indicative of hepatobiliary disease (Mosaad et al., 2003). Administration of HCE at different doses level (100 and 200 mg/kg) attenuated the increased
Table 1: Effect of HCE on serum AST (U/L), ALT (U/L), ALP (U/L), Bilirubin level (mg/dl), Albumin (gm/dl), Total protein (gm/dl), LDH (U/L), (mg/dl) and CHL (gm/dl) against R+I+P induced liver toxicity in rats

<table>
<thead>
<tr>
<th>Group</th>
<th>AST</th>
<th>ALT</th>
<th>ALP</th>
<th>TBL</th>
<th>ALB</th>
<th>TP</th>
<th>LDH</th>
<th>CHL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>96.28 ± 5.23</td>
<td>49.71 ± 4.02</td>
<td>62.23 ± 4.78</td>
<td>0.69 ± 0.15</td>
<td>4.78 ± 0.04</td>
<td>6.72 ± 0.07</td>
<td>425.31 ± 2.01</td>
<td>34.33 ± 2.3</td>
</tr>
<tr>
<td>R+I+P</td>
<td>378.68 ± 6.31†</td>
<td>239.25 ± 8.77†</td>
<td>157.57 ± 5.95†</td>
<td>3.21 ± 0.37†</td>
<td>3.11 ± 0.04†</td>
<td>3.9 ± 0.03†</td>
<td>721.21 ±3.1†</td>
<td>69.12±3.1†</td>
</tr>
<tr>
<td>HCE 100</td>
<td>358.61 ± 5.62a</td>
<td>218.41 ± 7.74a</td>
<td>136.69 ± 6.31a</td>
<td>2.39 ± 0.14b</td>
<td>3.13 ± 0.02n</td>
<td>4.13 ± 0.01a</td>
<td>711.12 ±3.0a</td>
<td>61.12 ±2.6a</td>
</tr>
<tr>
<td>HCE 200</td>
<td>120.37 ± 5.41c</td>
<td>87.27 ± 5.33c</td>
<td>89.65 ± 6.64c</td>
<td>0.83 ± 0.14c</td>
<td>3.29 ± 0.01b</td>
<td>5.41 ± 0.02c</td>
<td>616.21 ±3.0c</td>
<td>46.12 ±2.0c</td>
</tr>
<tr>
<td>Silymarin</td>
<td>108.88 ± 4.43c</td>
<td>58.84 ± 4.72c</td>
<td>71.34 ± 6.21c</td>
<td>0.71 ± 0.13c</td>
<td>4.28 ± 0.05c</td>
<td>6.22 ± 0.12c</td>
<td>554.23± 3.93c</td>
<td>38.12 ±1.8c</td>
</tr>
</tbody>
</table>

Values are mean ± S.E.M. of 6 rats in each group
n: non significant
P values: †<0.001 compared with respective control group I
P values:  a<0.05, b<0.01, c<0.001 compared with group II (R+I+P).

Figure legend 1.
Histopathology of liver tissues. (A) Liver section of normal control rat shows central vein surrounded by hepatic cord of cells (normal architecture). (B) Liver section of anti-tubercular drugs treated rats showing massive fatty changes, focal necrosis with portal inflammation, loss of cellular boundaries, central vein congestion and ballooning formation. (C) Liver section of rats treated anti-tubercular drugs and 100 mg/kg of HCE showing mild focal necrosis but massive fatty changes with mild central vein congestion, less inflammatory cells and less ballooning. (D) Liver section of rats treated anti-tubercular drugs and 200 mg/kg of HCE showing less inflammatory cells, absence of necrosis and congestion in central vein and showing regeneration of hepatocytes around central vein toward near normal liver architecture. (E) Liver section of rats treated anti-tubercular drugs and 100 mg/kg of silymarin showing normal liver architecture.
Table 2: Effect of HCE on antioxidant parameters like liver LPO (MDA nmole/min/mg of protein), SOD (unit/mg of protein), CAT (units/mg of protein) and GSH (nmole/mg of protein) against R+I+P induced liver toxicity in rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>LPO</th>
<th>GSH</th>
<th>SOD</th>
<th>CAT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.33 ± 0.08</td>
<td>0.86 ± 0.06</td>
<td>28.38 ± 2.63</td>
<td>61.61 ± 3.62</td>
</tr>
<tr>
<td>R+I+P</td>
<td>1.29 ± 0.11†</td>
<td>0.39 ± 0.03†</td>
<td>9.87 ± 1.62†</td>
<td>36.41 ± 2.48†</td>
</tr>
<tr>
<td>HCE 100</td>
<td>1.01 ± 0.09a</td>
<td>0.56 ± 0.05a</td>
<td>11.35 ± 1.21n</td>
<td>45.92 ± 2.56a</td>
</tr>
<tr>
<td>HCE 200</td>
<td>0.53 ± 0.06c</td>
<td>0.71 ± 0.04c</td>
<td>22.89 ± 2.61b</td>
<td>52.89 ± 3.85b</td>
</tr>
<tr>
<td>Silymarin</td>
<td>0.38 ± 0.07c</td>
<td>0.79 ± 0.06c</td>
<td>26.82 ± 2.72c</td>
<td>56.4 ± 2.51c</td>
</tr>
</tbody>
</table>

Values are mean ± S.E.M. of 6 rats in each group
n: non-significant
P values: †<0.001 compared with respective control group I
P values: a<0.05, b<0.01, c<0.001 compared with group II (R+I+P)

levels of the serum enzymes, produced by antitubercular drugs and caused a subsequent recovery towards normalization comparable to the control group I animals. The hepatoprotective effect of the HCE was further accomplished by the histopathological examinations. HCE at different dose levels offers hepatoprotection, but 200 mg/kg is more effective than lower dose. As an alternative to inducing cellular damage by covalent binding, there is evidence that these antitubercular drugs cause cellular damage through the induction of oxidative stress, a consequence of dysfunction of hepatic antioxidant defence system. The role of oxidative stress in the mechanism of antitubercular drugs induced hepatitis has been reported by (Attri et al. 2000).

The body has an effective defence mechanism to prevent and neutralize the free radical induced damage. This is proficient by a set of endogenous antioxidant enzymes such as SOD, and catalase. These enzymes constitute a mutually supportive team of defence against ROS (Amresh et al., 2007). The significantly reduced activities of SOD and CAT observed point out the hepatic damage in the rats administered with antitubercular drugs but on treatment with 100 and 200 mg/kg of HCE groups showed significant increase in the level of these enzymes which indicates the antioxidant activity of the Hedyotis corymbosa. Many antibiotics therapy can favour free radical production and causes cellular damage (Doroshow et al., 1982). The combination of antitubercular treatment (I+R+P) in experimental animals enhanced lipid peroxidation, indicating increased oxidative stress in liver (Skakun et al., 1992).

Increase in the level of lipid peroxides in liver reflected the hepatocellular damage. The depletion of antioxidant defences and/or raise in free radical production deteriorates the prooxidant-antioxidant balance, leading to oxidative stress-induced cell death. Depletion of reduced glutathione (GSH) is known to result in enhanced lipid peroxidation and excessive lipid peroxidation can cause increased glutathione consumption (Sodhi et al., 1997), as observed in the present study.

Furthermore, on treatment with different doses of HCE (100 and 200 mg/kg) significantly reduction in the level of lipid peroxidation an important cause of destruction and damage to hepatocellular membranes, and elevation in the level of GSH in liver. The increase in hepatic GSH level in the rats treated with HCE may be due to de novo GSH synthesis or GSH regeneration. On phytochemical screening, HCE has also been reported to contain oleanolic acid, ursolic acid and γ-sitosterol, ursolic acid exhibited potent hepatoprotective effects while oleanolic acid has been reported to increase the antioxidant components in the liver and it also increased maintained the hepatic glutathione, which plays an important role in protecting against RIP- induced liver injury.

Conclusion

From the results it is clear that ethanolic extract of the Hedyotis corymbosa has shown dose dependent activity among which at the dose level of 200 mg/kg, p.o. shows greater activity which is comparable with the control and standard groups and there by scientifically support its traditional use.

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