Results
4. Results

4.1 Studies on *Tinospora cordifolia*

The findings of the present investigation in liver, lung, kidney and forestomach of mice treated with hydroalcoholic extract of aerial roots of *Tinospora* and BHA are depicted in the Tables 1-6 and Figures 1-4. The body weight and body weight gain of mice remained unaffected by treatment with the extract and BHA.

4.1.1 Hepatic studies

The relative weights of liver were significantly increased (P < 0.05) in the animals treated with BHA and higher dose of *Tinospora*. The microsomal protein contents were significantly elevated following treatment with BHA (P < 0.05) as well as *Tinospora* (P < 0.005). Cytosolic protein level was found significantly elevated (P < 0.01) in BHA treated group as compared to that in control group (Table 1).

The microsomal fraction of liver homogenate was used for estimating the modulation of cytochrome P450 system (cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase) and lipid peroxidation. The cytosolic fraction was used for determining the activities of phase II enzymes (GST and DTD), antioxidative parameters (GSH, GPX, GR, SOD and CAT) and lactate dehydrogenase.

4.1.1.1 Cytochrome P450 system

The haemproteins, Cyt P450 and Cyt b5 were determined in the microsomal fraction of liver presented with the exception of cytochrome b5, a dose response related induction from their basal level relative to control group of animals (Group I). In animals treated with low and high doses of *Tinospora* extract (Group II & III), cytochrome P450 level was elevated 1.50 (P < 0.005) and 1.81 (P < 0.01) folds. However, in BHA fed animals (positive control, Group IV), cytochrome P450 level was comparable to that of control group (Group I). In contrast, cytochrome b5 level was significantly elevated around 1.28 folds in the positive control group (Group IV), in relation to the negative
control group (Group I). Furthermore, at higher dose level of treatment, *Tinospora* induced some increase in cytochrome b5 (P > 0.05) (Table 2).

The higher dose of *Tinospora* treatment caused a significant increase (P < 0.05) in the activities of both NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase whereas low dose caused significant increase (1.11 folds, P < 0.01) only in NADH-cytochrome b5 reductase over its control. BHA administration did not show any significant alteration in activities of both these enzymes as compared to their control values (Table 2).

### 4.1.1.2 Phase II enzymes

For evaluating the effect of *Tinospora* on phase II enzymes, glutathione S-transferase (GST) and DT-diaphorase (DTD) were assayed in the cytosol of liver homogenate. A clear dose dependent increase in the specific activity of hepatic GST, following extract treatment was evident. The specific activity of GST was 1.72 folds in Group II and 1.57 folds (P < 0.05) in Group III in comparison with that in control (Group I). Likewise, BHA fed animals also presented a similar trend in glutathione S-transferase specific activity, being maximally induced 2.98 folds (P < 0.005) relative to negative control value under identical experimental conditions. *Tinospora*, at the levels of treatment being currently examined, augmented the hepatic basal DT-diaphorase specific activity, as deduced from its determination only in cytosol fraction. The increase in the values were 1.30 folds (Group II, P < 0.005) and 1.45 folds (Group III, P < 0.005) relative to control group (Group I). In the positive control animals (Group IV), the enzyme level was increased 2.17 folds (P < 0.01) in comparison to that in negative control animals (Table 2).

### 4.1.1.3 Antioxidative parameters

The antioxidative parameters measured were reduced glutathione (GSH), glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase. The mice treated with *Tinospora* and BHA showed an increase in the level of reduced glutathione which was measured as acid soluble sulphydryl group. At low and high dose
levels of *Tinospora* treatment, the content of sulfhydryl group was elevated by 1.34 folds (Group II, $P < 0.005$) and 1.87 folds (Group III, $P < 0.005$) respectively. BHA treatment caused 2.17 folds ($P < 0.005$) increase in the content of sulfhydryl as compared to that in negative control animals. Only the low dose of treatment with the extract was effective in significantly elevating glutathione peroxidase and glutathione reductase activities, while it remained comparable to the control value with high dose of treatment. Furthermore, in positive control group (Group IV), only glutathione reductase was found significantly induced ($P < 0.005$) while glutathione peroxidase remain unaffected.

In case of SOD, except for a significant increase in the low dose treatment group by 1.25 folds (Group II; $P < 0.05$), none of the treatment groups exhibited any significant alterations in the specific activity of the enzyme. Treatment with low dose of *Tinospora* effectively increased the basal constitutive level of catalase by approximately 1.21 folds (Group II, $P < 0.05$), whereas with higher dose a further small increase in the enzyme level was noticed (Group III, $P < 0.05$). On the other hand, BHA treatment appreciably decreased the hepatic catalase activity (Group IV, $P < 0.01$). (Table 3).

### 4.1.1.4 Lipid peroxidation and lactate dehydrogenase

In the present study, lipid peroxidation estimated as MDA formation, was found to be effectively inhibited at both the levels of treatment with *Tinospora*, the order of potency being: high dose $>$ BHA $>$ low dose. Lower and higher dose of *Tinospora* produced about 31% ($P < 0.01$) and 40% ($P < 0.005$) decrease in lipid peroxidation respectively. BHA treated (positive control) animals also presented decrease in lipid peroxidation by 39% ($P < 0.01$) (Table 3). The specific activity of lactate dehydrogenase was significantly inhibited in animals treated with BHA (40%, $P < 0.005$) and higher dose of *Tinospora* (23%, $P < 0.005$) (Table 1).

### 4.1.2 Extrahepatic studies

The extrahepatic organs examined were lung, kidney and forestomach. The relative weights of these organs did not show any significant alteration following treatment with test material except in the lung of animals treated with lower dose of *Tinospora* (1.14...
folds, \( P < 0.05 \)). The protein contents in these extrahepatic organs of all experimental groups remained comparable to their respective control values (Tables 4-6).

The specific activities of GST, DTD, SOD and catalase were measured in post-mitochondrial supernatant fraction obtained after centrifuging the homogenate (10%, w/v) at 15,000 \( \times \) g for 30 min at 4°C, as described in the chapter on materials and methods.

### 4.1.2.1 Glutathione S-transferase

Only in lung and forestomach of animals exposed to *Tinospora*, glutathione S-transferase specific activity was significantly induced. At low dose of treatment, it was found maximally induced, the magnitude of elevation being 1.31 folds in lung (\( P < 0.01 \)); and 1.55 folds in forestomach (\( P < 0.01 \)). In BHA treated positive control animals (Group IV), the glutathione S-transferase specific activity was also increased significantly in all the extrahepatic organs examined viz. lung (\( P < 0.005 \)), kidney (\( P < 0.005 \)) and forestomach (\( P < 0.005 \)) (Table 4-6).

### 4.1.2.2 DT-diaphorase

Except for a significant alteration in the basal level of the activity induced by the extract in the forestomach, DT-diaphorase specific activity in lung and kidney of mice remained comparable with that of control groups. In forestomach of mice, a dose dependent increase in specific enzyme activity relative to control was evident. The enzyme activity increased by 1.07 folds (Group II, \( P < 0.05 \)) at lower dose level and 1.16 folds (Group III, \( P < 0.001 \)) at higher dose level over that of control group (Group I). In positive control group (Group IV), DT-diaphorase specific activity was significantly elevated in all the extrahepatic organs studied (Table 4-6) i.e., lung (\( P < 0.05 \)), kidney (\( P < 0.01 \)), and forestomach (\( P < 0.01 \)). Interestingly, in forestomach the magnitude of induction by BHA was comparatively low (1.13 folds, \( P < 0.01 \)) when compared to the level of induction elicited by treatment with high dose of *Tinospora* extract (1.17 folds, \( P < 0.005 \)).
4.1.2.3 Superoxide dismutase

The extrahepatic organs examined, revealed an increase in constitutive basal level of superoxide dismutase specific activity by *Tinospora* extract; however, only in lung the observed increase was seen to be dose related; whereas, in kidney and forestomach the lower dose rather than the higher dose appeared to be more potent in inducing the activity of enzyme. In lung, the magnitude of induction was: 1.22 folds at lower dose and 1.41 folds at the higher dose level; in kidneys, the sequence was - high dose (1.08 folds) < BHA (1.13 folds) < low dose (1.17 folds); and in forestomach the sequence was - high dose (1.23 folds) < low dose (1.33 folds) < positive control (1.79 folds). With exception in lung, BHA induced superoxide dismutase specific activity was above that of the basal level in forestomach and kidney (Group IV); in forestomach this presented the highest activity relative to control among all the treatment groups. In contrast, in kidney, BHA induced specific activity was less in magnitude as compared to group treated with the low dose of the extract. (Tables 4-6).

4.1.2.4 Catalase

Catalase activity detectable only in lung and kidney under our assay conditions, presented a significant alteration in the treated groups. In lung, it was maximally induced in group III by 1.42 folds (P < 0.005) relative to negative control, while in forestomach, maximal induction was 1.34 folds (Group III; P < 0.01) relative to negative control. Lower dose of *Tinospora* also caused a significant induction in lung (P < 0.01) and in kidney (P < 0.005). Furthermore, in lung and kidney, BHA induced catalase activity was above that induced by higher dose of *Tinospora*: (1.52 folds in lung: P < 0.01 and 1.43 folds in kidney: P < 0.005) (Tables 4-6).
Table 1. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on body weight gain and toxicity related parameters in mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt. x 100/Final body wt.</th>
<th>LDH ①</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle -d.w.)</td>
<td>27.5±1.41 (1.00)</td>
<td>28.5±0.926 (1.00)</td>
<td>4.45±0.362 (1.00)</td>
<td>1.83±0.181 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>28.3±1.28 (1.03)</td>
<td>29.3±1.04 (1.03)</td>
<td>4.61±0.234 (1.03)</td>
<td>1.75±0.149 (0.96)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>28.3±0.707 (1.03)</td>
<td>29.5±1.41 (1.03)</td>
<td>4.78±0.010 a (1.07)</td>
<td>1.41±0.124 c (0.77)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>26.5±0.926 (0.96)</td>
<td>27.3±1.04 (0.96)</td>
<td>4.69±0.450 a (1.05)</td>
<td>1.12±0.077 c (0.61)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in liver of mice receiving test substance to that of control mice).

a (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

①μ mole NADH oxidised/min/mg protein.

Abbreviations - BHA: butylated hydroxyanisole, d.w.: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 1. Effect of two doses of *Tinospora cordifolia* extract and BHA on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Tinospora*: 50 mg/kg body wt./day, Dose II of *Tinospora*: 100 mg/kg body wt./day and BHA: butylated hydroxyanisole 0.75% of diet.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 2. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 (1)</th>
<th>Cyt b5 (1)</th>
<th>Cyt P450 R (2)</th>
<th>Cyt b5 R (3)</th>
<th>GST (4)</th>
<th>DTD (5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.388±0.013 (1.00)</td>
<td>0.218±0.007 (1.00)</td>
<td>0.148±0.008 (1.00)</td>
<td>3.69±0.177 (1.00)</td>
<td>1.80±0.235 (1.00)</td>
<td>0.039±0.0029 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>0.576±0.037 1.50)</td>
<td>0.229±0.013 (1.04)</td>
<td>0.160±0.014 (1.08)</td>
<td>4.10±0.290 (1.11)</td>
<td>2.84±0.269 (1.57)</td>
<td>0.051±0.0038 (1.31)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>0.695±0.049 (1.81)</td>
<td>0.250±0.024 (1.19)</td>
<td>0.162±0.008 (1.10)</td>
<td>4.014±0.332 (1.12)</td>
<td>3.11±0.326 (1.72)</td>
<td>0.057±0.0047 (1.46)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>0.387±0.021 (1.00)</td>
<td>0.278±0.016 (1.28)</td>
<td>0.163±0.013 (1.10)</td>
<td>3.43±0.275 (0.93)</td>
<td>5.38±0.320 (2.99)</td>
<td>0.086±0.0105 (2.21)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in liver of mice receiving test substance to activity in liver of control mice).

\( ^a (p < 0.05), ^b (p < 0.01) \) and \( ^c (p < 0.005) \) represent significant changes against control.

**Abbreviations**- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: NADPH-cytochrome P450 reductase, Cyt b5 R: NADH-cytochrome b5 reductase, GST: glutathione S-transferase, DTD: DT-diaphorase and d.w.: distilled water.

Treatment duration: 14 days
Figure 2. Effect of two doses of *Tinospora cordifolia* extract and BHA on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Tinospora*: 50 mg/kg body wt/day, Dose II of *Tinospora*: 100 mg/kg body wt/day and BHA: butylated hydroxyanisole 0.75% of diet.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 3. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Sulphydryl content</th>
<th>GPX (nmole/min/mg protein)</th>
<th>GR (nmole NADPH consumed/min/mg protein)</th>
<th>SOD (units/mg protein)</th>
<th>CAT (nmole H2O2 consumed/min/mg protein)</th>
<th>LPO (nmole malondialdehyde formed/mg protein)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>25.4±2.53 (1.00)</td>
<td>52.3±4.56 (1.00)</td>
<td>35.6±4.16 (1.00)</td>
<td>6.28±0.663 (1.00)</td>
<td>83.1±3.58 (1.00)</td>
<td>0.627±0.040 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>33.9±3.28 (1.34)</td>
<td>61.9±3.82 (1.18)</td>
<td>56.2±5.36 (1.58)</td>
<td>7.87±0.623 (1.25)</td>
<td>100.8±7.12 (1.21)</td>
<td>0.431±0.024 (0.687)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>47.4±3.33 (1.87)</td>
<td>50.5±3.91 (0.97)</td>
<td>44.9±2.75 (1.26)</td>
<td>6.68±0.674 (1.06)</td>
<td>110.3±6.29 (1.32)</td>
<td>0.375±0.029 (0.598)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>55.2±4.44 (2.17)</td>
<td>50.3±4.01 (0.96)</td>
<td>52.0±4.61 (1.46)</td>
<td>6.16±0.376 (0.98)</td>
<td>59.1±3.77 (0.711)</td>
<td>0.382±0.029 (0.609)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in liver of mice receiving test substance to activity in liver of control mice).

* a (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

①nmole/g tissue, ②nmole of NADPH consumed/min/mg protein, ③specific activity is expressed as units/mg protein ④μmole H2O2 consumed/min/mg protein ⑤nmole malondialdehyde formed/mg protein.

Abbreviations- GPX: glutathione peroxidase, GR: glutathione reductase, SOD: superoxide dismutase, CAT: catalase, LPO: lipid peroxidation and d.w.: distilled water.

Treatment duration: 14 days
Figure 3. Effect of two doses of *Tinospora cordifolia* extract and BHA on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Tinospora*: 50 mg/kg body wt./day, Dose II of *Tinospora*: 100 mg/kg body wt./day and BHA: butylated hydroxyanisole 0.75% of diet.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 4. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on detoxifying and antioxidant enzymes profile in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST (1)</th>
<th>DTD (2)</th>
<th>SOD (3)</th>
<th>CAT (4)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.666±0.017 (1.00)</td>
<td>0.228±0.015 (1.00)</td>
<td>0.016±0.0004 (1.00)</td>
<td>5.18±0.266 (1.00)</td>
<td>20.6±2.28 (1.00)</td>
<td>7.14±0.615 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>0.697±0.063 (1.05)</td>
<td>0.300±0.026b (1.32)</td>
<td>0.016±0.0004 (1.00)</td>
<td>6.32±0.448c (1.22)</td>
<td>25.0±2.00b (1.21)</td>
<td>6.96±0.550 (0.98)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>0.762±0.066a (1.14)</td>
<td>0.272±0.024a (1.19)</td>
<td>0.016±0.0003 (1.00)</td>
<td>7.28±0.410c (1.41)</td>
<td>29.2±0.90c (1.42)</td>
<td>7.03±0.450 (0.98)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>0.696±0.049 (1.05)</td>
<td>0.334±0.020c (1.47)</td>
<td>0.018±0.0012c (1.13)</td>
<td>5.48±0.592 (1.06)</td>
<td>31.4±2.18b (1.52)</td>
<td>6.86±0.550 (0.96)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to activity in lung of control mice).

* (p < 0.05), ** (p < 0.01) and *** (p < 0.005) represent significant changes against control.

1) μmole CDNB-GSH conjugate formed/min/mg protein, 2) μmole DCPIP reduced/min/mg protein. 3) Specific activity is expressed as units/mg protein and 4) μmole H2O2 consumed/min/mg protein.

Abbreviations- GST: glutathione S-transferase, DTD: DT-diaphorase, SOD: superoxide dismutase, CAT: catalase and d.w.: distilled water.

Treatment duration: 14 days
Table 5. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on detoxifying and antioxidant enzymes profile in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST (@)</th>
<th>DTD (B)</th>
<th>SOD (C)</th>
<th>CAT (D)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>1.29±0.130 (1.00)</td>
<td>0.351±0.010 (1.00)</td>
<td>0.032±0.0012 (1.00)</td>
<td>7.27±0.180 (1.00)</td>
<td>8.50±0.290 (1.00)</td>
<td>7.03±0.386 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>1.32±0.094 (1.02)</td>
<td>0.330±0.025 (0.94)</td>
<td>0.032±0.0010 (1.00)</td>
<td>8.51±0.549b (1.17)</td>
<td>10.82±0.612c (1.27)</td>
<td>6.86±0.824 (0.98)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>1.30±0.116 (1.01)</td>
<td>0.379±0.024 (1.08)</td>
<td>0.033±0.0046 (1.03)</td>
<td>7.86±0.467a (1.08)</td>
<td>11.35±0.326b (1.34)</td>
<td>6.60±0.749 (0.94)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>1.19±0.079 (0.92)</td>
<td>0.441±0.024c (1.26)</td>
<td>0.048±0.0013b (1.50)</td>
<td>8.21±0.589c (1.13)</td>
<td>12.13±0.552c (1.43)</td>
<td>6.87±0.694 (0.98)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to kidney in liver of control mice).

a (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

@μmole CDNB-GSH conjugate formed/ min/mg protein, @μmole DCPIP reduced/ min/mg protein. @ specific activity is expressed as units/mg protein and @μmole H₂O₂ consumed/ min/mg protein.

Abbreviations- GST: glutathione S-transferase, DTD: DT-diaphorase, SOD: superoxide dismutase, CAT: catalase and d.w.: distilled water.

Treatment duration: 14 days
Table 6. Modulatory influence of two different doses of *Tinospora cordifolia* extract and BHA on detoxifying and antioxidant enzymes profile in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST ①</th>
<th>DTD &lt;br&gt;2</th>
<th>SOD &lt;br&gt;③</th>
<th>CAT &lt;br&gt;④</th>
<th>Protein &lt;br&gt;(mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>0.346±0.034 (1.00)</td>
<td>0.198±0.016 (1.00)</td>
<td>0.024±0.0008 (1.00)</td>
<td>5.10±0.434 (1.00)</td>
<td>ND</td>
<td>2.78±0.380 (1.00)</td>
</tr>
<tr>
<td></td>
<td>(only vehicle- d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Tinospora</em> (50 mg/kg body wt.)</td>
<td>0.331±0.025 (0.96)</td>
<td>0.309±0.024b (1.55)</td>
<td>0.026±0.0013a (1.08)</td>
<td>6.8±0.794c (1.33)</td>
<td>ND</td>
<td>3.02±0.256 (1.13)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Tinospora</em> (100 mg/kg body wt.)</td>
<td>0.392±0.018 (0.95)</td>
<td>0.267±0.019c (1.35)</td>
<td>0.028±0.0017c (1.17)</td>
<td>6.30±0.815c (1.23)</td>
<td>ND</td>
<td>3.04±0.300 (1.09)</td>
</tr>
<tr>
<td>Gr IV</td>
<td>BHA (0.75% in diet)</td>
<td>0.346±0.021 (1.00)</td>
<td>0.294±0.025e (1.49)</td>
<td>0.027±0.0013b (1.13)</td>
<td>9.15±0.990c (1.79)</td>
<td>ND</td>
<td>2.74±0.298 (0.99)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

① (p < 0.05), ② (p < 0.01) and ③ (p < 0.005) represent significant changes against control.

①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein, ③ specific activity is expressed as units/mg protein and ④μmole H₂O₂ consumed/min/mg protein.

Abbreviations- GST: glutathione S-transferase, DTD: DT-diaphorase, SOD: superoxide dismutase, CAT: catalase, d.w.: distilled water and ND: not detectable.

Treatment duration: 14 days
Figure 4. Effect of two doses of *Tinospora cordifolia* extract and BHA on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation.

Co: control, Dose I of *Tinospora*: 50 mg/kg body wt/day, Dose II of *Tinospora*: 100 mg/kg body wt/day and BHA: butylated hydroxyanisole 0.75% of diet. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Catalase activity in forestomach was not detectable.

Treatment duration: 14 days.
4.2 Studies on *Andrographis paniculata*

The results of the present investigation in liver, lung, kidney and forestomach of mice following treatment with hydroalcoholic extract of leaves and stem of *Andrographis* have been illustrated in Tables 7-12 and Figures 5-8. The body weight and body weight gain of mice treated with the plant extract remained unaltered as in case of untreated group of mice.

4.2.1 Hepatic Studies

Liver to final body weight ratio as well as microsomal and cytosolic protein contents did not show any significant change following treatment with the plant extract. The microsomal fraction of liver homogenate was used for estimating the modulation of cytochrome P450 system (cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase) and lipid peroxidation. The cytosolic fraction was used for determining the activities of phase II enzymes (GST and DTD), antioxidative parameters (GSH, GPX, GR, SOD and CAT) and lactate dehydrogenase.

4.2.1.1 Cytochrome P450 system

Microsomal haemproteins, cytochrome P450 and cytochrome b5, presented a contrasting pattern of inducing potential following *Andrographis* treatment. Cytochrome P450 at both doses revealed an induction of 1.21 and 1.38 folds relative to the control basal values, whereas cytochrome b5 in the treated animals exhibited a dose dependent decline in activity to the extent of 24% and 41% of the basal level of activity.

Compared to control, the specific activity of cytochrome P450 reductase was observed significantly elevated at both the dose levels investigated, being 1.10 folds in Group II and 1.15 folds in Group III. In contrast, specific activity of cytochrome b5 reductase presented a dose independent inducibility by *Andrographis* extract. At lower dose of treatment, its specific activity was induced by 20% (P < 0.01) whereas, with high dose of treatment it was elevated by 16% (P < 0.01) relative to the control value (Table 8).
4.2.1.2 Phase II enzymes

A significant dose dependent increase in hepatic cytosolic glutathione S-transferase specific activity relative to that in the control group of animals was evident. With lower dose of treatment, an induction of only 1.16 folds was observed as compared to that in Group I; whereas, higher dose of the modulator induced nearly 1.45 folds increase in glutathione S-transferase specific activity over that of the control value (Group III, $P < 0.001$).

DT-diaphorase specific activity evaluated only in cytosol of the treated animals also revealed a dose dependent increase in activity relative to control value (Group I). With lower and higher doses of treatment, the percentage increase in specific enzyme activity has been observed as 21% and 32 % respectively (Table 8).

Antioxidative parameters

The antioxidative parameters measured were reduced glutathione, glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase. The level of reduced glutathione measured as acid soluble sulfhydryl group showed an increase of 1.47 folds ($P < 0.005$) in Group II and 1.42 folds ($P < 0.01$) in Group III as compared to that in Group I. The specific activities of glutathione peroxidase and glutathione reductase were found significantly augmented with higher dose of the modulator treatment relative to their control levels ($P < 0.01$). At the lower dose of treatment, some induction, albeit statistically non-significant was evident. Compared to the value in control animals, specific activity of SOD has been found elevated to the extent of 32% ($P < 0.05$) in Group II and 53% ($P < 0.05$) in Group III, at the two different dose levels of extract being investigated. A statistically significant increase in catalase specific activity was discernable with higher dose of modulator treatment to the animals where, it was elevated by 21% ($P < 0.05$); at lower dose of treatment the increase was only 9 % which was not statistically significant (Table 9).
4.2.1.4 Lipid peroxidation and lactate dehydrogenase

*Andrographis* significantly inhibited the lipid peroxidation in a dose dependent manner. It was measured as the formation of malondialdehyde (MDA). The MDA formation was decreased by 40% ($P < 0.01$) in Group II and 44% ($P < 0.005$) in Group III as compared to that in untreated Group I. The specific activity of lactate dehydrogenase was also decreased following extract treatment but was found significant only with lower dose level of treatment (Group II, $P < 0.01$) (Table 7).

4.2.2 Extrahepatic organs

The extrahepatic organs examined were lung, kidney and forestomach. The relative weights of these organs did not show any significant alteration following *Andrographis* treatment except in the kidney of animals treated with its lower dose ($P < 0.05$). The protein levels in experimental groups of all the three organs, were comparable to their control values except in the lung of higher dose treated group and in the forestomach of lower dose treated group which showed a decrease of 10% ($P < 0.01$) and an increase of 10% ($P < 0.05$), respectively (Table 10-12).

The specific activities of GST, DTD, SOD and catalase were measured in post-mitochondrial supernatant fraction obtained after centrifuging the homogenate (10% w/v) at 15,000 $\times$ g for 30 minutes at 4°C as described in the chapter on materials and methods.

4.2.2.1 Glutathione S-transferase

With the exception of kidney of the experimental animals, no significant increase in GST specific activity was discernible in lung and forestomach. In kidney, an increase of 29% in Group II and 24% in Group III over that of control value have been observed by the lower and higher doses of *Andrographis* treatment respectively (Tables 10-12).

4.2.2.2 DT-diaphorase

DT-diaphorase specific activity as measured in post-mitochondrial supernatant revealed a significant increase in all the extrahepatic organs examined. Relative to that in control group, the specific enzyme activity was found maximally induced in kidney of the
Results

Experimental animals at both dose levels that being 1.32 folds (P < 0.01) in Group II and 1.27 folds (P < 0.01) in Group III over that of the control value. In lung, *Andrographis* resulted in increase in the specific enzyme activity by 1.13 folds (P < 0.001) in Group II and 1.12 folds (P < 0.05) in Group III. In forestomach of the experimental animals, induction of 15% (P < 0.01) in Group II and 15% (P < 0.001) in Group III, above the basal enzyme activity was noted (Tables 10-12).

4.2.2.3 Superoxide dismutase

The specific activity of superoxide dismutase as determined in lung, kidney and forestomach of the experimental animals revealed a significant induction above basal level of enzyme activity only in the lung and forestomach. In lung, a dose dependent increase in superoxide dismutase activity was noticed following *Andrographis* treatment to the animals. Compared to control value, an increase of 1.42 folds (P < 0.01) in Group II and 1.24 folds (P < 0.005) in Group III was evident. In the forestomach of both the treated groups of mice (Group II and Group III), an increase of 87% (P < 0.001) relative to that in control group was noted (Tables 10-12).

4.2.2.4 Catalase

In lung, the specific activity of catalase showed a dose dependent significant decline of 20% (P < 0.01) in Group II and 28% (P < 0.001) in Group III relative to control value. In contrast, in kidney of the experimental animals, the specific activity of catalase was seen significantly increased by lower dose of treatment with *Andrographis* extract (Group II, P < 0.01). In forestomach of the control and experimental animals, no catalase specific activity was detectable under our assay conditions (Tables 10-12).
Table 7. Modulatory influence of two different doses of *Andrographis paniculata* leaf extract on body weight gain and toxicity related parameters in mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt x 100/ Final body wt.</th>
<th>LDH (1)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial Final</td>
<td></td>
<td></td>
<td>Microsome Cytosol</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>22.0±1.51 (1.00) 23.0±1.51 (1.00)</td>
<td>4.29±0.378 (1.00)</td>
<td>2.80±0.194 (1.00)</td>
<td>11.02±1.108 (1.00) 9.52±0.680 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td>22.0±1.07 (1.00) 23.3±1.04 (1.01)</td>
<td>4.57±0.550 (1.06)</td>
<td>2.30±0.272 (0.82)</td>
<td>11.35±1.111 (1.03) 10.05±1.126 (1.05)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Andrographis</em> (100 mg/kg body wt.)</td>
<td>21.4±1.51 (0.97) 22.5±1.41 (0.98)</td>
<td>4.30±0.217 (1.00)</td>
<td>2.71±0.320 (0.97)</td>
<td>11.88±1.178 (1.08) 9.53±0.931 (1.00)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

\( ^b (p < 0.01) \) represents significant change against control.

\( ^\text{1}\mu\text{mole/mg protein.} \)

Abbreviation- d.w.: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 5. Effect of two doses of *Andrographis paniculata* leaf extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice.

Error bars represent standard deviation.

Co: control, Dose I of *Andrographis*: 50 mg/kg body wt./day and Dose II of *Andrographis*: 100 mg/kg body wt./day.

b(P < 0.01) indicates significant change against control.

Treatment duration: 14 days.
Table 8. Modulatory influence of two different doses of *Andographis paniculata* leaf extract on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450</th>
<th>Cyt b5</th>
<th>Cyt P450 R</th>
<th>Cyt b5 R</th>
<th>GST</th>
<th>DTD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.580±0.034 (1.00)</td>
<td>0.210±0.014 (1.00)</td>
<td>0.145±0.007 (1.00)</td>
<td>3.53±0.231 (1.00)</td>
<td>2.32±0.152 (1.00)</td>
<td>0.019±0.0020 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td>0.700±0.071c (1.21)</td>
<td>0.160±0.015b (0.76)</td>
<td>0.160±0.009a (1.10)</td>
<td>4.24±0.287b (1.20)</td>
<td>2.69±0.270a (1.16)</td>
<td>0.023±0.0020b (1.21)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Andrographis</em> (100 mg/kg body wt)</td>
<td>0.801±0.079c (1.38)</td>
<td>0.124±0.012b (0.59)</td>
<td>0.166±0.016a (1.15)</td>
<td>4.09±0.291a (1.16)</td>
<td>3.36±0.161c (1.45)</td>
<td>0.025±0.0015c (1.32)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

* (p < 0.05), † (p < 0.01) and ‡ (p < 0.005) represent significant changes against control.

1μmole/mg protein, 2μmole of NADPH oxidised/min/mg protein, 3μmole of NADH oxidised/min/mg protein 4μmole CDNB-GSH conjugate formed/min/mg protein and 5μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 6. Effect two doses of *Andrographis paniculata* on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5), and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-Cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Andrographis*: 50 mg/kg body wt./day and Dose II of *Andrographis*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 9. Modulatory influence of two different doses of *Andrographis paniculata* on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>GSH</th>
<th>GPX</th>
<th>GR</th>
<th>SOD</th>
<th>CAT</th>
<th>LPO</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control (only vehicle-d.w.)</td>
<td>40.9±3.22</td>
<td>62.7±4.24</td>
<td>41.1±4.24</td>
<td>5.24±0.521</td>
<td>28.0±2.80</td>
<td>0.519±0.053</td>
</tr>
<tr>
<td></td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td>60.0±7.90(^c)</td>
<td>69.7±5.75</td>
<td>46.0±2.97</td>
<td>6.94±0.653(^b)</td>
<td>30.5±2.89</td>
<td>0.312±0.030(^b)</td>
</tr>
<tr>
<td></td>
<td><em>Andrographis</em> (100 mg/kg body wt.)</td>
<td>57.9±4.90(^b)</td>
<td>78.6±3.26(^c)</td>
<td>51.9±3.90(^a)</td>
<td>8.02±0.583(^c)</td>
<td>33.9±2.81(^a)</td>
<td>0.292±0.014(^c)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

\(^a\) (p < 0.05), \(^b\) (p < 0.01) and \(^c\) (p < 0.005) represent significant changes against control.

\(^1\)nmole GSH/g tissue, \(^2\)nmole of NADPH consumed/min/mg protein, \(^3\)specific activity expressed as \(\mu\)mole/mg protein, \(^4\)\(\mu\)mole \(\text{H}_2\text{O}_2\) consumed/min/mg protein and \(^5\)nmole malondialdehyde formed/mg protein.


Treatment duration: 14 days
Figure 7: Effect of two doses of *Andrographis paniculata* leaf extract on reduced glutathione content (GSH), specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Andrographis*: 50 mg/kg body wt./day and Dose II of *Andrographis*: 100 mg/kg body wt./day. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control. Treatment duration: 14 days.
Table 10. Modulatory influence of two different doses of *Andrographis paniculata* on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST (1)</th>
<th>DTD (2)</th>
<th>SOD (3)</th>
<th>CAT (4)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td></td>
<td>0.730±0.513  (1.00)</td>
<td>0.329±0.024  (1.00)</td>
<td>0.0252±0.0016 (1.00)</td>
<td>5.83±0.257 (1.00)</td>
<td>31.10±3.53 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td></td>
<td>0.743±0.468  (1.02)</td>
<td>0.366±0.034  (1.11)</td>
<td>0.0285±0.0013 (1.13)</td>
<td>8.30±0.717b (1.42)</td>
<td>24.90±2.51b (0.80)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Andrographis</em> (100 mg/kg body wt.)</td>
<td></td>
<td>0.797±0.599  (1.09)</td>
<td>0.335±0.022  (1.02)</td>
<td>0.0283±0.0019a (1.12)</td>
<td>7.25±0.593c (1.24)</td>
<td>22.5±1.55c (0.72)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lungs of mice receiving test substance to activity in lungs of control mice).

* (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

1 μmole CDNB-GSH conjugate formed/min/mg protein, 2 μmole DCPIP reduced/min/mg protein 3 specific activity expressed as μmole/mg protein and 4 μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 11. Modulatory influence of two different doses of *Andrographis paniculata* on detoxifying and antioxidant enzymes profile in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>1.10±0.110 (1.00)</td>
<td>0.254±0.038 (1.00)</td>
<td>0.0414±0.0019 (1.00)</td>
<td>5.43±0.496 (1.00)</td>
<td>13.74±1.170 (1.00)</td>
<td>5.74±0.485 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td>1.23±0.078a (1.12)</td>
<td>0.328±0.029b (1.29)</td>
<td>0.054±0.0040b (1.32)</td>
<td>6.25±0.544 (1.15)</td>
<td>15.8±0.631b (1.15)</td>
<td>5.85±0.745 (1.02)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Andrographis</em> (100 mg/kg body wt.)</td>
<td>1.29±0.123a (1.17)</td>
<td>0.316±0.026a (1.24)</td>
<td>0.052±0.0018b (1.27)</td>
<td>6.08±0.423 (1.12)</td>
<td>14.1±0.403 (1.03)</td>
<td>5.70±0.555 (1.01)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to activity in lungs of control mice).

* a (p < 0.05) and b (p < 0.01) represent significant changes against control.

①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein ③specific activity expressed as μmole/mg protein and ④μmole H$_2$O$_2$ consumed/min/mg protein.


Treatment duration: 14 days
Table 12. Modulatory influence of two different doses of *Andrographis paniculata* on detoxifying and antioxidant enzymes profile in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.228±0.042 (1.00)</td>
<td>0.120±0.017 (1.00)</td>
<td>0.0220±0.0008 (1.00)</td>
<td>2.06±0.233 ND</td>
<td>8.29±0.568 (1.00)</td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Andrographis</em> (50 mg/kg body wt.)</td>
<td>0.256±0.025 (1.12)</td>
<td>0.153±0.020 ①</td>
<td>0.0252±0.0628 ②</td>
<td>3.85±0.425 ③</td>
<td>ND</td>
<td>8.41±0.263 (1.01)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Andrographis</em> (100 mg/kg body wt.)</td>
<td>0.238±0.019 (1.04)</td>
<td>0.132±0.013 ①</td>
<td>0.0253±0.0010 ②</td>
<td>3.85±0.325 ③</td>
<td>ND</td>
<td>8.45±0.305 (1.02)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in lungs of control mice).

① (p < 0.05), ② (p < 0.01) and ③ (p < 0.005) represent significant changes against control.

①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein ③specific activity expressed as μmole/mg protein and ④μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Figure 8. Effect of two doses of *Andrographis paniculata* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice.

Error bars represent standard deviation.

Co: control, Dose I of *Andrographis*: 50 mg/kg body wt/day and Dose II of *Andrographis*: 100 mg/kg body wt/day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Catalase activity in forestomach was not detectable.

Treatment duration: 14 days.
4.3 Studies on *Adhatoda vesica*

The results of the present investigation are depicted in Tables 13-18 and Figures 9-12. The treatment with hydroalcoholic extract of *Adhatoda* leaf resulted in increase of body weight and body weight gain of mice though it was not significant.

4.3.1 Hepatic Studies

Liver to final body weight ratio showed a significant increase at both dose levels of treatment; 1.14 folds (P < 0.05) in Group II and 1.15 folds (P < 0.005) in Group III. Increase in microsomal and cytosolic protein contents were seen in both treated groups. However, it was significant only in cytosolic protein, 1.16 folds (P < 0.05) in Group II and 1.23 folds (P < 0.005) in Group III.

The microsomal fraction of liver homogenate was used for estimating the modulation of cytochrome P450 system (cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase) and lipid peroxidation. The cytosolic fraction was used for determining the activities of phase II enzymes (GST and DTD), antioxidative parameters (GSH, GPx, GR, SOD and catalase) and lactate dehydrogenase.

4.3.1.1 Cytochrome P450 system

The components of cytochrome P450 system were determined in microsomal fraction of liver. Cytochrome P450 and cytochrome b5 showed significant increase in their contents at both dose levels of treatment. Cytochrome P450 presented a significant elevation of 17% in Group II and 16% in Group III. Cytochrome b5 showed a dose dependent induction in *Adhatoda* treated animals, which was 1.12 folds (P < 0.05) in Group II and 1.33 folds (P < 0.01) in Group III as compared to that in control group.

Cytochrome P450 reductase and cytochrome b5 reductase also showed significant increase (P < 0.005) above their basal level specific activities, at both doses of treatment. The specific activity of cytochrome P450 reductase was increased by 1.34 and 1.28 folds in lower and higher dose treated groups respectively. Cytochrome b5 reductase showed a
dose dependent induction which was 1.17 folds in Group II and 1.19 folds in Group III as compared to control value (Group I).

4.3.1.2 Phase II enzymes

For studying the effect of *Adhatoda* on drug metabolizing phase II enzymes, the specific activities of glutathione S-transferase (GST) and DT-diaphorase (DTD) were measured. Extract treatment to experimental groups of mice induced a distinct dose dependent increase in the activity of hepatic GST, which was 17% (P < 0.05) in lower dose treated mice and 35% (P < 0.01) in higher dose treated mice over that of control. Specific activity of cytosolic DTD was also significantly augmented in both *Adhatoda* treated groups (Group II and Group III), though maximum increase of 1.20 folds (P < 0.001) was seen at lower dose level of treatment (Table 15).

4.3.1.3 Antioxidative parameters

Reduced glutathione measured as acid soluble sulfhydryl group (-SH) in liver homogenate presented significant elevation at both dose levels of *Adhatoda* treatment. The elevated levels were 1.82 folds (P < 0.005) in Group II and 1.70 folds (P < 0.005) in Group III as compared to that in control. The other antioxidative parameters measured were glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD) and catalase which also presented significant increase at both dose levels of extract treatment. GPX, SOD and catalase showed dose dependent increase of 1.24, 1.15 and 2.25 folds respectively in Group II; and 1.28, 1.38 and 2.80 folds respectively in Group III. The specific activity of GR was elevated by 19% (P < 0.01) in Group II and 7% (P < 0.05) in Group III over that of control value (Table 15).

4.3.1.4 Lipid peroxidation and Lactate dehydrogenase

Lipid peroxidation estimated as MDA formation in the microsomal fraction of liver homogenate, was significantly (P < 0.005) inhibited by almost 30% in both extract treated groups. Administration of *Adhatoda* lowered the activity of lactate dehydrogenase but it was not significant when compared with control value.
4.3.2 Extrahepatic organs

The relative weights of lung, kidney and forestomach did not show any significant alteration following *Adhatoda* treatment except in lower dose treated group of kidney (P < 0.05). The protein levels in experimental groups of all the three organs, were comparable to their control values except in the lung of higher dose treated mice and in the forestomach of lower dose treated mice, which showed a decrease of 10% (P < 0.01) and an increase of 10% (P < 0.05) respectively (Tables 16-18).

The specific activities of GST, DTD, SOD and catalase were measured in post-mitochondrial supernatant fraction obtained after centrifuging the homogenate (10% w/v) at 15,000 × g for 30 minutes at 4°C as described in the chapter on materials and methods.

4.3.2.1 Glutathione S-transferase

All the extrahepatic organs examined, showed increase in the specific activity of glutathione S-transferase in both extract treated groups as compared to their respective control values. The induction in the activity of GST was significant as well as dose-dependent in lung and forestomach. In lower dose treated mice, the GST activity in lung and forestomach was elevated by 1.30 and 1.18 folds (P < 0.005) respectively; in higher dose treated mice it was elevated by 1.38 and 1.30 folds (P < 0.001) respectively. In kidney, though GST activity was increased by 13% in Group II and 11% in Group III, it was not significant relative to the control value. (Tables 16-18).

4.3.2.2 DT-diaphorase

The mice treated with *Adhatoda*, revealed an increase in the basal level activity of DT-diaphorase in lung, kidney and in forestomach. A dose dependent modulation in specific activity of DTD was evident in lung, which was 1.18 folds (P < 0.005) in Group II and 1.29 folds (P < 0.001) in Group III. Kidney showed significant increase in DTD activity at higher dose level by 7% (P < 0.05) and forestomach at lower dose level by 19% (P < 0.05) relative to their control values (Tables 16-18).
4.3.2.3 Superoxide dismutase

The specific activity of superoxide dismutase in kidney increased significantly in animals treated with lower and higher dose of extract; the increases were 66% and 39% (P < 0.005) respectively. In contrast, lung showed a decrease of about 10% (P < 0.05) in both the extract treated groups of mice. In forestomach, SOD activity was induced slightly but was not significant as compared to its control value (Tables 16-18).

4.3.2.4 Catalase

Catalase activity, detectable only in the supernatants of lung and kidney under our assay conditions, showed significant alterations following Adhatoda treatment. In lung, catalase activity was significantly elevated by 1.19 folds (P < 0.01) at lower dose level of treatment whereas in kidney the significant increase of 1.28 folds (P < 0.005) was found at higher dose level of treatment (Tables 16-17).
Table 13. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on body weight gain and toxicity related parameters in mouse.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt. x100/Final body wt.</th>
<th>LDH (μM/mg)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td>Microsome</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>26.5±0.93 (1.00)</td>
<td>27.8±1.98 (1.00)</td>
<td>5.42±0.452 (1.00)</td>
<td>1.45±0.156 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>26.8±1.04 (1.01)</td>
<td>29.3±2.12 (1.05)</td>
<td>6.18±0.535 (1.14)</td>
<td>1.29±0.190 (0.89)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>27.5±0.93 (1.04)</td>
<td>29.8±2.71 (1.07)</td>
<td>6.25±0.378 (1.15)</td>
<td>1.29±0.136 (0.89)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

* (p < 0.05) and † (p < 0.005) represent significant changes against control.

†μmole/mg protein.

Abbreviation - d.w.: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 9. Effect of two doses of *Adhatoda vesica* leaf extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Adhatoda*: 50 mg/kg body wt./day and Dose II of *Adhatoda*: 100 mg/kg body wt./day.

a(P < 0.05) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 14. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 ① (nmole/mg protein)</th>
<th>Cyt b5 ① (μM of NADPH oxidised/min/mg protein)</th>
<th>Cyt P450 R ② (μM of NADH oxidised/min/mg protein)</th>
<th>Cyt b5 R ③ (μM of CDNB-GSH conjugate formed/min/mg protein)</th>
<th>GST ④ (μM of DCPIP reduced/min/mg protein)</th>
<th>DTD ⑤ (μM of DCPIP reduced/min/mg protein)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.457±0.014 (1.00)</td>
<td>0.305±0.019 (1.00)</td>
<td>0.215±0.020 (1.00)</td>
<td>3.71±0.255 (1.00)</td>
<td>2.39±0.198 (1.00)</td>
<td>0.0156±0.0005 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>0.536±0.028b (1.17)</td>
<td>0.341±0.016a (1.12)</td>
<td>0.289±0.018c (1.34)</td>
<td>4.34±0.251c (1.17)</td>
<td>2.79±0.257a (1.17)</td>
<td>0.0183±0.0010d (1.17)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>0.528±0.048a (1.16)</td>
<td>0.404±0.024b (1.33)</td>
<td>0.275±0.013c (1.28)</td>
<td>4.41±0.214c (1.19)</td>
<td>3.23±0.20b (1.55)</td>
<td>0.0173±0.0014a (1.11)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

* (p < 0.05), b (p < 0.01), c (p < 0.005) and d (p < 0.001) represent significant changes against control.

①nmole/mg protein, ②μmole of NADPH oxidised/min/mg protein, ③μmole of NADH oxidised/min/mg protein ④μmole CDNB-GSH conjugate formed/min/mg protein and ⑤μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 10. Effect of two doses of *Adhatoda vesica* leaf extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice.

Error bars represent standard deviation.

Co: control, Dose I of *Adhatoda*: 50 mg/kg body wt./day and Dose II of *Adhatoda*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control.

Treatment duration: 14 days.
Table 15. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>GSH  (1)</th>
<th>GPX  (2)</th>
<th>GR  (2)</th>
<th>SOD  (3)</th>
<th>CAT  (4)</th>
<th>LPO  (5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>27.3±2.70</td>
<td>76.2±2.41</td>
<td>53.3±2.51</td>
<td>5.61±0.490</td>
<td>52.4±6.11</td>
<td>0.675±0.021</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>50.8±4.25 (1.82)</td>
<td>94.7±3.39 (1.24)</td>
<td>63.6±2.27 (1.19)</td>
<td>6.45±0.290 (1.15)</td>
<td>117.7±11.08 (2.25)</td>
<td>0.473±0.037 (0.70)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>46.4±3.53 (1.70)</td>
<td>97.4±3.74 (1.28)</td>
<td>56.9±1.65 (1.07)</td>
<td>7.75±0.252 (1.38)</td>
<td>146.8±16.68 (2.80)</td>
<td>0.490±0.029 (0.73)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

\( ^a (p < 0.05), ^b (p < 0.01) \) and \( ^c (p < 0.005) \) represent significant changes against control.

\( 1 \text{n mole GSH/g tissue, 2 n mole of NADPH consumed/min/mg protein, 3 specific activity expressed as } \mu \text{mole/mg protein, 4 } \mu \text{mole H}_2\text{O}_2 \text{ consumed/min/mg protein and 5 nmole malondialdehyde formed/mg protein.} \)


Treatment duration: 14 days
Figure 11. Effect of two doses of *Adhatoda vesica* leaf extract on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Adhatoda*: 50 mg/kg body wt./day and Dose II of *Adhatoda*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 16. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST (μmole/min/mg protein)</th>
<th>DTD (μmole/min/mg protein)</th>
<th>SOD (μmole/mg protein)</th>
<th>CAT (μmole/min/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.536±0.048 (1.00)</td>
<td>0.209±0.023 (1.00)</td>
<td>0.017±0.0010 (1.00)</td>
<td>4.01±0.419 (1.00)</td>
<td>47.2±4.38 (1.00)</td>
<td>3.55±0.169 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>0.605±0.059 (1.13)</td>
<td>0.271±0.019 (1.30)</td>
<td>0.020±0.0012 (1.18)</td>
<td>3.24±0.409 (0.81)</td>
<td>56.1±4.44 (1.19)</td>
<td>3.36±0.224 (0.95)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>0.589±0.064 (1.10)</td>
<td>0.289±0.003 (1.38)</td>
<td>0.022±0.0010 (1.29)</td>
<td>3.15±0.237 (0.79)</td>
<td>50.8±5.17 (1.08)</td>
<td>3.21±0.232 (0.90)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to activity in lung of control mice).

*(p < 0.05), **(p < 0.01), *** (p < 0.005) and **** (p < 0.001) represent significant changes against control.*

Specific activity expressed as: 1μmole CDNB-GSH conjugate formed/min/mg protein, 2μmole DCPIP reduced/min/mg protein 3specific activity expressed as μmole/mg protein and 4μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 17. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST 1</th>
<th>DTD 2</th>
<th>SOD 3</th>
<th>CAT 4</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>1.39±0.53 (1.00)</td>
<td>0.228±0.030 (1.00)</td>
<td>0.015±0.0008 (1.00)</td>
<td>3.44±0.233 (1.00)</td>
<td>84.85±7.86 (1.00)</td>
<td>6.21±0.56 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>1.47±0.10 (1.06)</td>
<td>0.258±0.029 (1.13)</td>
<td>0.015±0.0008 (1.00)</td>
<td>5.73±0.436 (1.66)</td>
<td>85.22±9.71 (1.00)</td>
<td>5.98±0.59 (0.96)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>1.43±0.136 (1.03)</td>
<td>0.254±0.020 (1.11)</td>
<td>0.016±0.0010* (1.07)</td>
<td>4.78±0.443* (1.39)</td>
<td>108.71±9.06* (1.28)</td>
<td>6.07±0.57 (0.98)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to activity in kidney of control mice).

* (p < 0.05) and ** (p < 0.005) represent significant changes against control.

Specific activity expressed as: 1μmole CDNB-GSH conjugate formed/min/mg protein, 2μmole DCPIP reduced/min/mg protein 3specific activity expressed as μmole/mg protein and 4μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 18. Modulatory influence of two different doses of *Adhatoda vesica* leaf extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.192±0.022 (1.00)</td>
<td>0.528±0.050 (1.00)</td>
<td>0.026±0.0023 (1.00)</td>
<td>3.65±0.359</td>
<td>ND</td>
<td>4.11±0.354 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Adhatoda</em> (50 mg/kg body wt.)</td>
<td>0.189±0.025 (0.98)</td>
<td>0.623±0.041c (1.18)</td>
<td>0.031±0.0032a (1.19)</td>
<td>3.92±0.540</td>
<td>ND</td>
<td>4.52±0.233a (1.10)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Adhatoda</em> (100 mg/kg body wt.)</td>
<td>0.185±0.014 (0.96)</td>
<td>0.691±0.041d (1.30)</td>
<td>0.028±0.0034 (1.08)</td>
<td>4.06±0.548</td>
<td>ND</td>
<td>4.36±0.433 (1.06)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

① (p < 0.05), ② (p < 0.01) and ③ (p < 0.005) represent significant changes against control.

Specific activity expressed as: ①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein ③specific activity expressed as μmole/mg protein and ④μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Figure 12. Effect of two doses of *Adhatoda vesica* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation.

Co: control, Dose I of *Adhatoda*: 50 mg/kg body wt/day and Dose II of *Adhatoda*: 100 mg/kg body wt/day.

a*(P < 0.05)*, b*(P < 0.01)*, c*(P < 0.005)* and d*(P < 0.001)* indicate significant changes against control.

Catalase activity in forestomach was not detectable.
4.4 Studies on *Aloe vera*

The findings of the present study have been depicted in Tables 19-24 and figures 13-16.

The experimental groups of mice treated with *Aloe* leaf extract showed significant increase in body weight and body weight gain at both the dose levels of treatment (Table 19).

4.4.1 Hepatic studies

*Aloe* treated groups of mice showed slight increase in liver-somatic index as compared to that in control group. Protein levels were significantly increased in microsomal as well as cytosolic fractions in both the extract treated groups. In lower dose treated group, protein levels were increased by 13% \((P < 0.05)\) and 37% \((P < 0.001)\) whereas in higher dose treated group the increase in protein levels were 10% \((P < 0.05)\) and 20% \((P < 0.01)\) in microsome and cytosol respectively (Table 19, Figure 13).

4.4.1.1 Cytochrome P450 system

The estimated levels of cytochrome P450 and cytochrome b5 presented statistically significant decrease in both *Aloe* treated groups as compared to their controls except Group III of Cyt b5 in which the decrease (9%) in the level was not found significant. The maximum reduction of 38% \((P < 0.005)\) was evident in Cyt P450 at high dose level of treatment. In lower dose treated group of mice, levels of Cyt P450 and Cyt b5 were reduced by 13% \((P < 0.05)\) and 8% \((P < 0.05)\) respectively.

A dose dependent significant increase was observed in specific activities of NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase at both the dose levels of treatment relative to their control values. The percentage of elevation in Cyt P450 reductase and Cyt b5 reductase was 9% and 14% in low dose treated group; 22% and 28% in high dose treated group respectively (Table 20, Figure 14).
4.4.1.2 Phase II enzymes

In phase II enzymes, GST and DTD were assayed in the cytosol of liver homogenate. Hepatic glutathione S-transferase showed a significant increase of 1.19 folds \((P < 0.005)\) in its specific activity in higher dose treated group of mice whereas in lower dose treated group it was comparable to that in control. Hepatic DT-diaphorase showed a dose dependent induction in its specific activity following *Aloe* treatment. The activity of DTD was elevated by 1.15 \((P < 0.05)\) and 1.22 \((P < 0.005)\) folds respectively at lower and higher dose levels of treatment as compared to the values in the control group (Table 20, Figure 14).

4.4.1.3 Antioxidative parameters

The antioxidant parameters studied were GSH, GPx, GR, SOD and catalase. The level of reduced glutathione (GSH) measured as acid soluble sulfhydryl group (-SH) was significantly elevated in a dose dependent manner following *Aloe* treatment. The magnitude of elevation in the -SH level was 1.37 folds \((P < 0.005)\) in Group II and 1.42 folds \((P < 0.005)\) Group III as compared to that in Group I. Only higher dose of *Aloe* treatment was effective in significantly elevating the specific activities of glutathione peroxidase (GPX) and glutathione reductase (GR) while it remained comparable to control values following treatment with lower dose. The activities of GPX and GR were elevated respectively by 1.25 folds \((P < 0.05)\) and 1.30 \((P < 0.005)\) folds in higher dose treated group as compared to the values in their control mice. Superoxide dismutase and catalase presented a statistically significant as well as dose dependent induction in their activities at both the dose levels of treatment. The magnitude of induction of SOD and catalase was 1.09 folds \((P < 0.05)\) and 1.91 folds \((P < 0.01)\) in Group II; 1.44 folds \((P < 0.005)\) and 2.12 folds \((P < 0.005)\) in Group III respectively over that of their control values. (Table 21, Figure 15).

4.4.1.4 Lipid peroxidation and lactate dehydrogenase

The extent of lipid peroxidation estimated as MDA formation, was significantly inhibited by 20% \((P < 0.01)\) at higher dose level of treatment. In lower dose treated mice
it was reduced by 8% but was not significant as compared to the control value (Table 21). The specific activity of LDH was reduced in a dose dependent manner following *Aloe* treatment. The magnitude of reduction was 53 % (*P* < 0.01) and 55% (*P* < 0.005) in Group II and Group III respectively a compared to that in control group (Table 19).

### 4.4.2 Extrahepatic studies

There were no significant alterations in the relative weights of extrahepatic organs (lung, kidney and forestomach) examined, following *Aloe* treatment. The level of protein also did not show any significant change in kidney and forestomach over that of their controls. In lung, a significant increase of 1.12 folds (*P* < 0.01) in protein content was evident at lower dose level of treatment (Tables 22-24).

#### 4.4.2.1 Glutathione S-transferase

The extrahepatic organs examined, revealed a dose dependent increase in constitutive basal level of specific activity of glutathione S-transferase, following administration of *Aloe*. The magnitude of change was in the order of lung > kidney > forestomach at both the dose levels of treatment (Group II and Group III) as compared to that in their controls. In lower dose treated group, the elevation in the GST activity was 1.25 folds (*P* < 0.001), 1.15 folds (*P* < 0.05) and 1.11 folds (*P* < 0.05) in lung, kidney and forestomach respectively. In higher dose treated group, the increase was found to be 1.33 folds (*P* < 0.001), 1.30 folds (*P* < 0.001) and 1.18 folds (*P* < 0.01) in lung, kidney and forestomach respectively (Tables 22-24, Figure 16).

#### 4.4.2.2 DT-diaphorase

*Aloe* treated groups of mice presented an increase in the specific activity of DT-diaphorase in all the extrahepatic organs examined, but was found significant only in the forestomach of Group II (1.17 folds, *P* < 0.001), and in forestomach and kidney of Group III (1.25 folds, *P* < 0.005; 1.15 folds, *P* < 0.05 respectively) (Tables 22-24, Figure 16).
4.4.2.3 Superoxide dismutase

The specific activity of superoxide dismutase in lung, kidney and forestomach was significantly elevated in all the Aloe treated groups of mice. However, the maximal induction observed in lower dose treated group was 1.37 folds, 1.55 folds and 1.21 folds in lung, kidney and forestomach, respectively as compared to their control values. The higher dose treated group showed increase in SOD activity by 31%, 14% and 16% in lung, kidney and forestomach respectively (Tables 22-24, Figure 16).

4.4.2.4 Catalase

Catalase activity was detectable only in lung and kidney supernatants under our present assay conditions. It presented significant alterations following Aloe treatment. In lower dose treated group, specific activity of catalase was increased by 35% (P < 0.005) in lung and 29% (P < 0.001) in kidney; and in higher dose treated group it was increased by 18% (P < 0.005) in lung and 40% (P < 0.001) in kidney (Tables 22-24, Figure 16).
Table 19. Modulatory influence of two different doses of *Aloe vera* leaf extract on body weight gain and toxicity related parameters in mouse.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt.x100/ Final body wt.</th>
<th>LDH (unit/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial Final</td>
<td></td>
<td>Microsome Cytosol</td>
<td></td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>21.25±1.49 (1.00)</td>
<td>22.00±1.85 (1.00)</td>
<td>5.28±0.337 (1.00)</td>
<td>2.89±0.288 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aloe</em> (30 µl/ animal)</td>
<td>22.00±1.07 (1.04)</td>
<td>25.25±1.83b (1.15)</td>
<td>5.45±0.396 (1.03)</td>
<td>1.36±0.223b (0.47)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aloe</em> (60 µl/ animal)</td>
<td>22.25±1.04 (1.05)</td>
<td>26.75±2.12c (1.22)</td>
<td>5.79±0.516 (1.10)</td>
<td>1.30±0.172c (0.45)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

* (p < 0.05),  b (p < 0.01),  c (p < 0.005) and  d (p < 0.001) represent significant changes against control.

μmole/mg protein.

Abbreviation- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 13. Effect of two doses of Aloe vera leaf extract on liversomatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of Aloe: 30 µl/animal/day and Dose II of Aloe: 60 µl/animal/day. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control. Treatment duration: 14 days.
Table 20. Modulatory influence of two different doses of *Aloe vera* leaf extract on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 ①</th>
<th>Cyt b5 ①</th>
<th>Cyt P450 R ②</th>
<th>Cyt b5 R ③</th>
<th>GST ④</th>
<th>DTD ⑤</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>0.600 ± 0.046 (1.00)</td>
<td>0.229 ± 0.009 (1.00)</td>
<td>0.242 ± 0.013 (1.00)</td>
<td>3.78 ± 0.191 (1.00)</td>
<td>1.82 ± 0.181 (1.00)</td>
<td>0.027 ± 0.0017 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aloe</em> (30μl/animal)</td>
<td>0.522 ± 0.034a (0.87)</td>
<td>0.210 ± 0.012a (0.92)</td>
<td>0.263 ± 0.012a (1.09)</td>
<td>4.32 ± 0.169b (1.14)</td>
<td>1.84 ± 0.120 (1.01)</td>
<td>0.031 ± 0.0030a (1.15)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aloe</em> (60μl/animal)</td>
<td>0.373 ± 0.042c (0.62)</td>
<td>0.209 ± 0.019 (0.91)</td>
<td>0.296 ± 0.022c (1.22)</td>
<td>4.83 ± 0.198c (1.28)</td>
<td>2.17 ± 0.149c (1.19)</td>
<td>0.033 ± 0.0012c (1.22)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

a (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

① nmole/mg protein, ② μmole of NADPH oxidised/min/mg protein, ③ μmole of NADH oxidised/min/mg protein
④ μmole CDNB-GSH conjugate formed/min/mg protein and ⑤ μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 14. Effect of two doses of Aloe vera leaf extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5), and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of Aloe: 30μl/animal/day and Dose II of Aloe: 60μl/animal/day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 21. Modulatory influence of two different doses of *Aloe vera* leaf extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>GSH (1)</th>
<th>GPX (2)</th>
<th>GR (2)</th>
<th>SOD (3)</th>
<th>CAT (4)</th>
<th>LPO (5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>25.10±2.83 (1.00)</td>
<td>61.02±2.94 (1.00)</td>
<td>47.4±3.77 (1.00)</td>
<td>6.13±0.377 (1.00)</td>
<td>76.10±5.76 (1.00)</td>
<td>0.521±0.037 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aloe</em> (30μl/animal)</td>
<td>34.30±3.94 (1.37)</td>
<td>61.26±4.31 (1.00)</td>
<td>47.7±2.56 (1.01)</td>
<td>6.66±0.445 (1.09)</td>
<td>145.50±12.64 (1.91)</td>
<td>0.479±0.030 (0.92)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aloe</em> (60μl/animal)</td>
<td>35.70±4.88 (1.42)</td>
<td>76.41±5.07 (1.25)</td>
<td>56.8±3.03 (1.20)</td>
<td>8.83±0.475 (1.44)</td>
<td>161.9±13.61 (2.12)</td>
<td>0.415±0.035 (0.80)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

* (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

(1)nmole GSH/g tissue, (2)nmole of NADPH consumed/min/mg protein, (3)specific activity expressed as μmole/mg protein, (4)μmole H₂O₂ consumed/min/mg protein and (5)nmole malondialdehyde formed/mg protein.


Treatment duration: 14 days
Figure 15. Effect of two doses of \textit{Aloe vera} leaf extract on the reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice.

Error bars represent standard deviation.

Co: control, Dose I of \textit{Aloe}: 30 µl/animal/day and Dose II of \textit{Aloe}: 60 µl/animal/day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 22. Modulatory influence of two different doses of *Aloe vera* leaf extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST (μmol)</th>
<th>DTD (μmol)</th>
<th>SOD (μmol)</th>
<th>CAT (μmol)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>(Gr)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.758±0.064 (1.00)</td>
<td>0.150±0.015 (1.00)</td>
<td>0.012±0.0008 (1.00)</td>
<td>3.11±0.429 (1.00)</td>
<td>17.99±1.93 (1.00)</td>
<td>4.17±0.183 (1.00)</td>
</tr>
<tr>
<td>Gr II Aloe (30 μl/animal)</td>
<td>0.759±0.069 (1.00)</td>
<td>0.188±0.010d (1.25)</td>
<td>0.012±0.0008 (1.00)</td>
<td>4.25±0.471b (1.37)</td>
<td>24.32±1.81e (1.35)</td>
<td>4.65±0.195b (1.12)</td>
<td></td>
</tr>
<tr>
<td>Gr III Aloe (60 μl/animal)</td>
<td>0.808±0.066 (1.07)</td>
<td>0.199±0.018d (1.33)</td>
<td>0.013±0.0009 (1.08)</td>
<td>4.08±0.418a (1.31)</td>
<td>21.15±1.41e (1.18)</td>
<td>4.43±0.218 (1.06)</td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to lung of control mice).

* (p < 0.05), ** (p < 0.01), *** (p < 0.005) and **** (p < 0.001) represent significant changes against control.

1 μmol CDNB-GSH conjugate formed/min/mg protein, 2 μmol DCPIP reduced/min/mg protein 3 Specific activity expressed as μmol/mg protein and 4 μmol H2O2 consumed/min/mg protein.


Treatment duration: 14 days
Table 23. Modulatory influence of two different doses of *Aloe vera* leaf extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST 1</th>
<th>DTD 2</th>
<th>SOD 3</th>
<th>CAT 4</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>1.37±0.108 (1.00)</td>
<td>0.128±0.016 (1.00)</td>
<td>0.013±0.0008 (1.00)</td>
<td>3.72±0.389 (1.00)</td>
<td>57.43±5.18 (1.00)</td>
<td>5.67±0.254 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aloe</em> (30 µl/animal)</td>
<td>1.38±0.102 (1.01)</td>
<td>0.147±0.011 a (1.15)</td>
<td>0.014±0.0007 (1.08)</td>
<td>5.75±0.506 d (1.55)</td>
<td>74.18±6.03 d (1.29)</td>
<td>5.82±0.482 (1.03)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aloe</em> (60 µl/animal)</td>
<td>1.39±0.134 (1.02)</td>
<td>0.166±0.016 d (1.30)</td>
<td>0.015±0.0010 a (1.15)</td>
<td>4.23±0.253 a (1.14)</td>
<td>80.15±5.69 d (1.40)</td>
<td>5.68±0.384 (1.00)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to kidney of control mice).

* (p < 0.05) and ** (p < 0.001) represent significant changes against control.

1 µmole CDNB-GSH conjugate formed/min/mg protein, 2 µmole DCPIP reduced/min/mg protein
3 Specific activity expressed as µmole/mg protein and 4 µmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 24. Modulatory influence of two different doses of Aloe vera leaf extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST (1)</th>
<th>DTD (2)</th>
<th>SOD (3)</th>
<th>CAT (4)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.319±0.022 (1.00)</td>
<td>0.198±0.019 (1.00)</td>
<td>0.012±0.0008 (1.00)</td>
<td>3.53±0.429 (1.00)</td>
<td>ND 5.41±0.491 (1.00)</td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td>Aloe (30 µl/animal)</td>
<td>0.307±0.032 (0.96)</td>
<td>0.220±0.023a (1.11)</td>
<td>0.014±0.0008d (1.17)</td>
<td>4.26±0.330b (1.21)</td>
<td>ND 5.47±0.486 (1.01)</td>
<td></td>
</tr>
<tr>
<td>Gr III</td>
<td>Aloe (60 µl/animal)</td>
<td>0.308±0.029 (0.97)</td>
<td>0.233±0.018b (1.18)</td>
<td>0.015±0.0009c (1.25)</td>
<td>4.08±0.391a (1.16)</td>
<td>ND 5.12±0.597 (0.95)</td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

\( ^a (p < 0.05) \), \( ^b (p < 0.01) \), \( ^c (p < 0.005) \) and \( ^d (p < 0.001) \) represent significant changes against control.


Treatment duration: 14 days
Figure 16. Effect of two doses of *Aloe vera* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation. Co: control, Dose I of *Aloe*: 30 μl/animal/day and Dose II of *Aloe*: 60 μl/animal/day. a(P < 0.05), b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control. Catalase activity in forestomach was not detectable. Treatment duration: 14 days.
4.5 Studies on *Aegle marmelos*

The findings of present investigation are depicted in Tables 25-30 and Figures 17-20. The mice treated with hydroalcoholic extract of *Aegle* leaf did not show any significant alterations in body weight and body weight gain as compared to the control group of mice.

4.5.1 Hepatic studies

There was no significant change in liver-somatic index in *Aegle* treated mice. The protein levels in microsomal and cytosolic fractions of liver, also did not show any significant change following extract treatment. (Table 25).

4.5.1.1 Cytochrome P450 system

Different components of cytochrome P450 system were determined in microsomal fraction of liver. The haemproteins, cytochrome P450 and cytochrome b5 showed significant increase at lower dose level of treatment as compared to their control values. The level of induction at lower dose level was 1.15 folds ($P < 0.01$) in Cyt P450 and 1.12 folds ($P < 0.05$) in Cyt b5 (Table 26, Figure 18).

*Aegle* treated groups (Group II and Group III) presented significant elevation in the specific activities of NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase as compared to their controls. In case of Cyt P450 reductase, the elevation in activity was 1.23 folds ($P < 0.005$) in Group II and 1.32 folds ($P < 0.005$) in Group II showing a dose dependent response. The corresponding increase in the activity of Cyt b5 reductase were 1.22 ($P < 0.001$) and 1.15 ($P < 0.05$) folds against control value (Table 26, Figure 18).

4.5.1.2 Phase II enzymes

In phase II enzymes, glutathione S-transferase and DT-diaphorase were assayed in the cytosol of liver. Both these enzymes showed a significant dose dependent increase in their activities in *Aegle* treated groups of mice. At lower dose level of treatment specific
activities of GST and DTD were elevated by 44% ($P < 0.005$) and 14% ($P < 0.05$) and at higher dose level of treatment, the activities were increased by 66% ($P < 0.005$) and 20% ($P < 0.01$), respectively over the values in respective controls (Table 26, Figure 18).

4.5.1.3 Antioxidative parameters

Antioxidative parameters viz. reduced glutathione, glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase measured, showed dose response related increase in their level/activities, following *Aegle* treatment. Level of reduced glutathione in liver homogenate, measured as acid soluble sulphydryl group(-SH) was increased by 1.34 folds ($P < 0.005$) in Group II and 1.41 folds ($P < 0.005$) in Group III as compared to control level. At lower dose level of treatment, the specific activities of GPX, GR, SOD and catalase were significantly increased by 1.32, 1.24, 1.21 and 1.30 folds respectively; at higher dose level of treatment the significant increases were 1.45, 1.27, 1.37 and 1.35 folds respectively in comparison to their control values (Table 27, Figure 19).

4.5.1.4 Lipid peroxidation and lactate dehydrogenase

MDA formation was taken as an indicator of lipid peroxidation (LP). Lipid peroxidation and specific activity of lactate dehydrogenase both showed a dose dependent decrease following the administration of *Aegle*. At lower dose level of treatment, LP and specific activity of LDH were reduced by 27% ($P < 0.005$) and 26% ($P < 0.005$) respectively; and at higher dose level of treatment the values were reduced by 40% ($P < 0.005$) and 34% ($P < 0.005$) respectively as compared to their control values (Tables 25 and 27).

4.5.2 Extrahepatic studies

The relative weights of extrahepatic organs examined viz. lung, kidney and forestomach remained unaffected following treatment of *Aegle*. The protein contents generally showed a decreasing trend in *Aegle* treated groups. It was significantly reduced
Results in kidney in lower dose treated group; and in forestomach in groups treated with lower and higher doses of *Aegle* (Tables 28-30).

4.5.2.1 Glutathione S-transferase

The treatment with *Aegle* leaf extract showed a significant as well as dose dependent increase in the specific activity of GST in kidney and forestomach relative to their control values. Lung also showed a slight increase in its activity but was not significant as compared to the control value. In kidney and forestomach GST activity was elevated by 16% (P < 0.05) and 24% (P < 0.01) at lower dose level of treatment; and by 24% (P < 0.005) and 32% (P < 0.005) at higher dose level of treatment respectively (Tables 28-30, Figure 20).

4.5.2.2 DT-diaphorase

Like GST, the specific activity of DTD also presented an increase at both dose levels of extract treatment in lung, kidney and forestomach, but was found significant only at higher dose level of treatment as compared to respective control values. DTD activity in Group III (100 mg/kg body wt.) was elevated by 1.14 (P < 0.05), 1.20 (P < 0.01) and 1.14 (P < 0.05) folds in lung, kidney and forestomach respectively (Tables 28-30, Figure 20).

4.5.2.3 Superoxide dismutase

In kidney, extract treatment resulted into decrease in the activity of SOD by 39% (P < 0.005) in Group II and by 31% (P < 0.005) in Group III as compared to that in control group. In contrast, forestomach showed significant enhancement at both dose levels of *Clerodendrum* treatment that was 1.19 folds (P < 0.01) in Group II and 1.15 folds (P < 0.05) in Group III relative to its control value. The specific activity of SOD did not show any significant increase in lung (Tables 28-30, Figure 20).

4.5.2.4 Catalase

Under our assay conditions, the catalase activity was detectable only in lung and kidney. It showed significant as well as dose dependent increase only in kidney following
*Aegle* treatment. Catalase activity in kidney was enhanced by 1.37 folds ($P < 0.005$) in Group II and 1.61 folds ($P < 0.005$) in Group III). In lung, mice treated with low dose of extract showed 12% increase in catalase activity over that of control, but it was not significant (Tables 28-29, Figure 20).
Table 25. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on body weight gain and toxicity related parameters.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt.x 100/Final body wt.</th>
<th>LDH (µmole/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td>Microsome</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>27.3±1.04 (1.00)</td>
<td>28.0±1.16 (1.00)</td>
<td>5.44±0.523 (1.00)</td>
<td>3.15±0.428 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em> (50 mg/kg body wt.)</td>
<td>26.3±0.76 (0.96)</td>
<td>26.8±1.04 (0.96)</td>
<td>5.10±0.476 (0.94)</td>
<td>2.32±0.370 (0.74)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em> (100 mg/ kg body wt.)</td>
<td>26.8±1.04 (0.98)</td>
<td>28.5±0.93 (1.02)</td>
<td>5.82±0.420 (1.07)</td>
<td>2.07±0.333 (0.66)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

*(p <0.005)* represents significant change against control.

1µmole/mg protein.

Abbreviation- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 17. Effect of two doses of *Aegle marmelos* leaf extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Aegle*: 50 mg/kg body wt./day and Dose II of *Aegle*: 100 mg/kg body wt./day.

c(P < 0.005) indicates significant change against control.

Treatment duration: 14 days.
Table 26. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450</th>
<th>Cyt b5</th>
<th>Cyt P450 R</th>
<th>Cyt b5 R</th>
<th>GST</th>
<th>DTD</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>①</td>
<td>②</td>
<td>③</td>
<td>④</td>
<td>⑤</td>
<td></td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.470±0.015 (1.00)</td>
<td>0.183±0.008 (1.00)</td>
<td>0.377±0.036 (1.00)</td>
<td>3.12±0.250 (1.00)</td>
<td>4.56±0.157 (1.00)</td>
<td>0.021±0.0019 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em> (50 mg/kg body wt.)</td>
<td>0.543±0.024 ② (1.15)</td>
<td>0.204±0.016 ② (1.12)</td>
<td>0.463±0.033② (1.23)</td>
<td>3.81±0.214② (1.22)</td>
<td>6.57±0.508② (1.44)</td>
<td>0.024±0.0015② (1.14)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em> (100 mg/kg body wt.)</td>
<td>0.423±0.042 ② (0.90)</td>
<td>0.171±0.011 ② (0.93)</td>
<td>0.497±0.024② (1.32)</td>
<td>3.60±0.325② (1.15)</td>
<td>7.59±0.438② (1.66)</td>
<td>0.025±0.0015② (1.20)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

① (p < 0.05), ② (p < 0.01), ③ (p < 0.005) and ④ (p < 0.001) represent significant changes against control.

① μmole/mg protein, ② μmole of NADPH oxidised/min/mg protein, ③ μmole of NADH oxidised/min/mg protein
④ μmole CDNB-GSH conjugate formed/min/mg protein and ⑤ μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 18. Effect of two doses of *Aegle marmelos* leaf extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Aegle*: 50 mg/kg body wt./day and Dose II of *Aegle*: 100 mg/kg body wt./day. a(P < 0.05), b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control. Treatment duration: 14 days.
Table 27. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatment</th>
<th>GSH</th>
<th>GPX</th>
<th>GR</th>
<th>SOD</th>
<th>CAT</th>
<th>LPO</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>(Gr)</td>
<td>(Gr)</td>
<td>(Gr)</td>
<td>(Gr)</td>
<td>(Gr)</td>
<td>(Gr)</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>29.5±2.90</td>
<td>62.0±3.18</td>
<td>34.2±2.66</td>
<td>10.1±0.626</td>
<td>47.0±3.66</td>
<td>1.095±0.134</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em> (50 mg/kg body wt.)</td>
<td>39.7±4.26</td>
<td>82.0±6.02</td>
<td>42.4±3.32</td>
<td>12.2±0.986</td>
<td>61.2±3.88</td>
<td>0.803±0.069</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em> (100 mg/kg body wt.)</td>
<td>41.6±4.31</td>
<td>90.02±5.34</td>
<td>43.7±3.29</td>
<td>13.8±1.242</td>
<td>63.6±3.60</td>
<td>0.655±0.861</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

b (p < 0.01) and c (p < 0.005) represent significant changes against control.

1 nmole GSH/g tissue, 2 nmole of NADPH consumed/min/mg protein, 3 specific activity expressed as μmole/mg protein, 4 μmole H₂O₂ consumed/min/mg protein and 5 nmole malondialdehyde formed/mg protein.


Treatment duration: 14 days
Figure 19. Effect of two doses of *Aegle marmelos* leaf extract on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Aegle*: 50 mg/kg body wt./day and Dose II of *Aegle*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 28. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.612±0.053 (1.00)</td>
<td>0.288±0.029 (1.00)</td>
<td>0.014±0.0007 (1.00)</td>
<td>2.99±0.431 (1.00)</td>
<td>21.7±3.29 (1.00)</td>
<td>3.93±0.400 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em> (50 mg/kg body wt.)</td>
<td>0.608±0.037 (0.99)</td>
<td>0.301±0.036 (1.05)</td>
<td>0.015±0.0014 (1.07)</td>
<td>4.20±0.594b (1.41)</td>
<td>24.4±3.17 (1.12)</td>
<td>3.92±0.324 (1.00)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em> (100 mg/kg body wt.)</td>
<td>0.665±0.058 (1.09)</td>
<td>0.305±0.032 (1.06)</td>
<td>0.016±0.0008a (1.14)</td>
<td>4.01±0.630a (1.34)</td>
<td>21.4±3.07 (0.99)</td>
<td>3.59±0.339 (0.91)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to activity in lung of control mice).

① (p < 0.05) and ② (p < 0.01) represent significant changes against control.

①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein ③specific activity expressed as μmole/mg protein and ④μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 29. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST ¹</th>
<th>DTD ²</th>
<th>SOD ³</th>
<th>CAT ⁴</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>1.09±0.042 (1.00)</td>
<td>0.263±0.029 (1.00)</td>
<td>0.02±0.0011 (1.00)</td>
<td>5.95±0.508 (1.00)</td>
<td>48.6±3.80 (1.00)</td>
<td>5.34±0.409 (1.00)</td>
</tr>
<tr>
<td></td>
<td>(only vehicle-d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em></td>
<td>1.05±0.075 (0.96)</td>
<td>0.306±0.037⁵ (1.16)</td>
<td>0.021±0.0011 (1.05)</td>
<td>7.69±0.421⁵ (1.30)</td>
<td>66.5±6.31⁵ (1.37)</td>
<td>4.92±0.32⁵ (0.92)</td>
</tr>
<tr>
<td></td>
<td>(50 mg/kg body wt.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em></td>
<td>1.10±0.094 (1.01)</td>
<td>0.325±0.024⁶ (1.24)</td>
<td>0.024±0.0022b (1.20)</td>
<td>7.41±0.589⁶ (1.25)</td>
<td>78.1±7.08⁶ (1.61)</td>
<td>5.01±0.547 (0.94)</td>
</tr>
<tr>
<td></td>
<td>(100 mg/kg body wt.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to activity in kidney of control mice).

* (p < 0.05), ᵇ (p < 0.01) and ᶜ (p < 0.005) represent significant changes against control.

¹ μmole CDNB-GSH conjugate formed/min/mg protein, ²μmole DCPIP reduced/min/mg protein, ³specific activity expressed as μmole/mg protein and ⁴μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 30. Modulatory influence of two different doses of *Aegle marmelos* leaf extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST 1</th>
<th>DTD 2</th>
<th>SOD 3</th>
<th>CAT 4</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.171±0.016 (1.00)</td>
<td>0.451±0.045 (1.00)</td>
<td>0.014±0.0014 (1.00)</td>
<td>3.87±0.406 (1.00)</td>
<td>ND</td>
<td>5.64±0.320 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Aegle</em> (50 mg/kg body wt.)</td>
<td>0.167±0.023 (0.98)</td>
<td>0.560±0.052 b (1.24)</td>
<td>0.015±0.0014 (1.07)</td>
<td>6.64±0.546 c (1.72)</td>
<td>ND</td>
<td>4.98±0.286 b (0.88)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Aegle</em> (100 mg/kg body wt.)</td>
<td>0.187±0.018 (1.09)</td>
<td>0.595±0.050 c (1.32)</td>
<td>0.016±0.0016 a (1.14)</td>
<td>5.58±0.347 c (1.44)</td>
<td>ND</td>
<td>5.21±0.368 a (0.91)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

* (p < 0.05), ** (p < 0.01) and *** (p < 0.005) represent significant changes against control.

1 μmole CDNB-GSH conjugate formed/min/mg protein, 2 μmole DCPIP reduced/min/mg protein
3 Specific activity expressed as μmole/mg protein and 4 μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Figure 20. Effect of two doses of *Aegle marmelos* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation. Co: control, Dose I of *Aegle*: 50 mg/kg body wt/day and Dose II of *Aegle*: 100 mg/kg body wt/day. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control. Catalase activity in forestomach was not detectable. Treatment duration: 14 days
4.6 Studies on *Clerodendrum inerme*

The findings of the experiment carried out with *Clerodendrum* leaf extract are illustrated in Tables 31-36 and Figures 21-24. The treatment with extract did not affect the body weight and body weight gain in mice.

4.6.1 Hepatic studies

Mice treated with only the lower dose (Group II, 50 mg/kg body wt.) of the test material presented a significant increase in the relative weight of liver whereas those treated with higher dose (Group III, 100 mg/kg body wt.) matched the control value. The levels of protein in microsomal as well as cytosolic fractions of extract treated groups were found to be comparable to their control values except in microsome of higher dose treated group, which showed a significant elevation \((P < 0.01)\) in the protein level against that of control (Table 31).

4.6.1.1 Cytochrome P450 system

The main components of cytochrome P450 system are cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase. The levels of Cyt P450 and Cyt b5 were significantly elevated at both dose levels of extract treatment, but it was dose dependent only in case of Cyt b5. At lower dose level of treatment, the contents of Cyt P450 and Cyt b5 were increased by 1.19 \((P < 0.005)\) folds and 1.28 \((P < 0.005)\) folds; and at higher dose level by 1.12 \((P < 0.05)\) folds and 1.89 \((P < 0.001)\) folds, respectively as compared to their respective control values.

The specific activity of Cyt P450 reductase showed an increase of 1.18 folds \((P < 0.01)\) at lower dose level of treatment as compared to the control value. The other values in extract treated groups of Cyt P450 reductase and Cyt b5 reductase did not show any significant alteration relative to their respective control values (Table 32, Figure 22).
4.6.1.2 Phase II enzymes

To analyze the effect of *Clerodendrum* leaf extract on phase II enzymes, glutathione S-transferase and DT-diaphorase were assayed. Both of these enzymes showed a distinct dose response related enhancement in their specific activities following treatment with the test substance. In lower dose treated mice, activities of GST and DTD were elevated by 1.36 ($P < 0.005$) and 1.20 ($P < 0.05$) folds; and in higher dose treated mice by 1.62 ($P < 0.005$) and 1.53 ($P < 0.005$) folds, respectively as compared to their respective control values (Table 32, Figure 22).

4.6.1.3 Antioxidative parameters

All antioxidative enzymes measured, presented a significant as well as dose dependent increase in their activities following *Clerodendron* treatment. The content of reduced glutathione measured as acid soluble sulfhydryl group (-SH) in liver homogenate, increased by 2.12 ($P < 0.005$) folds in Group II and 1.74 ($P < 0.001$) folds in Group III as compared to that in control.

At lower dose level of treatment the specific activities of glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase were significantly increased by 1.32 ($P < 0.01$), 1.27 ($P < 0.01$), 1.32 ($P < 0.005$) and 1.17 ($P < 0.01$) folds respectively; and in higher dose by 1.82 ($P < 0.01$), 1.36 ($P < 0.01$), 1.91 ($P < 0.005$) and 1.33 ($P < 0.01$) folds respectively over that of their control values (Table 33, Figure 23).

4.6.1.4 Lipid peroxidation and lactate dehydrogenase

The extent of lipid peroxidation, as estimated from the formation of MDA, was found inhibited at both dose levels of *Clerodendron* treatment, but significant inhibition (27%, $P < 0.005$) was shown only in higher dose treated group of mice as compared to that in control (Table 33, Figure 23). Lactate dehydrogenase activity was inhibited in both Group II and Group III, but it was not significant when compared to control value (Table 31, Figure 23).
4.6.2 Extrahepatic studies

The relative weights of extrahepatic organs examined viz. lung, kidney and forestomach were comparable to their control values except in lung which showed a significant increase of 12% \((P < 0.05)\) in higher dose treated group.

In lung, the protein level was significantly elevated in both the extract treated groups i.e. 1.07 folds \((P < 0.05)\) in Group II and 1.13 folds \((P < 0.005)\) in Group III whereas in kidney, it was unaltered. In case of forestomach it was found significantly elevated only in high dose treated mice (Group III, 1.14 folds, \(P < 0.05\)) against control value (Tables 34-36).

4.6.2.1 Glutathione S-transferase

In lung, GST presented a significant as well as dose dependent increase at both lower and higher dose levels which were 1.28 folds \((P < 0.001)\) and 1.58 folds \((P < 0.001)\) respectively (Table 34, Figure 24). In kidney, the increase in GST activity was significant only at higher dose level of treatment (Group III; 1.17 folds, \(P < 0.005\)) whereas in forestomach, the values were found almost unaltered (Tables 34-36).

4.6.2.2 DT-diaphorase

In lung and forestomach there was no significant alteration in the specific activity of DTD following Clerodendrum treatment. Only kidney showed a significant as well as dose dependent increase in the activity of DTD in extract treated groups. The activity of DTD in kidney was elevated by 1.20 \((P < 0.05)\) folds and 1.26 \((P < 0.001)\) folds at lower and higher dose levels respectively as compared to control values (Tables 34-36, Figure 24).

4.6.2.3 Superoxide dismutase

Specific activity of SOD in lung was not affected by Clerodendrum treatment. Kidney showed a significant increase of 1.15 folds \((P < 0.05)\) in SOD activity at higher dose level of treatment whereas in forestomach it was found significant at low dose level.
of treatment (1.19 folds, \( P < 0.01 \)) as compared to their in control values (Tables 34-36, Figure 24).

4.6.2.4 Catalase

Under our assay conditions, catalase activity was detectable only in lung and kidney. It showed a significant as well as dose dependent increase in kidney. In this organ, the catalase activity was increased by 1.29 folds (\( P < 0.01 \)) in Group II and 1.39 folds (\( P < 0.01 \)) in Group III. In lung, specific activity of catalase was induced by 1.42 folds (\( P < 0.005 \)) and 1.38 folds (\( P < 0.005 \)) at lower and higher dose levels of treatment, respectively (Tables 34-35, Figure 24).
Table 31. Modulatory influence of two different doses of *Clerodendrum inerme* leaf extract on body weight gain and toxicity related parameters.

<table>
<thead>
<tr>
<th>Group (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt.x100/ Final body wt.</th>
<th>LDH (μmole/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td>Microsome</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>26.5±1.41 (1.00)</td>
<td>27.5±1.77 (1.00)</td>
<td>4.48±0.375 (1.00)</td>
<td>1.96±0.174 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Clerodendrum</em> (50 mg/kg body wt.)</td>
<td>26.8±1.04 (1.01)</td>
<td>28.5±0.96 (1.04)</td>
<td>5.01±0.400a (1.12)</td>
<td>1.77±0.200 (0.90)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Clerodendrum</em> (100 mg/kg body wt.)</td>
<td>27.5±0.93 (1.04)</td>
<td>30.5±0.93 (1.11)</td>
<td>4.50±0.342 (1.01)</td>
<td>1.84±0.151 (0.94)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

a (*p* < 0.05) and b (*p* < 0.01) represent significant changes against control.

Abbreviations- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 21. Effect of two doses of *Clerodendrum inerme* leaf extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Clerodendrum*: 50 mg/kg body wt./day and Dose II of *Clerodendrum*: 100 mg/kg body wt./day.
a(P < 0.05) and b(P < 0.01) indicate significant changes against control.

Treatment duration: 14 days.
Table 32. Modulatory influence of two different doses of *Clerodendrum inerme* on hepatic phase I and phase II related enzymes in mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 (N)</th>
<th>Cyt b5 (N)</th>
<th>Cyt P450 R (N)</th>
<th>Cyt b5 R (N)</th>
<th>GST (N)</th>
<th>DTD (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>0.577±0.060 (1.00)</td>
<td>0.123±0.009 (1.00)</td>
<td>0.209±0.009 (1.00)</td>
<td>3.57±0.138 (1.00)</td>
<td>2.93±0.158 (1.00)</td>
<td>0.015±0.0009 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Clerodendrum</em> (50 mg/kg body wt.)</td>
<td>0.685±0.037 (1.19)</td>
<td>0.157±0.015 (1.28)</td>
<td>0.247±0.028 (1.18)</td>
<td>3.98±0.430 (1.11)</td>
<td>3.99±0.227 (1.36)</td>
<td>0.018±0.0017 (1.20)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Clerodendrum</em> (100 mg/kg body wt.)</td>
<td>0.646±0.036 (1.12)</td>
<td>0.233±0.022 (1.89)</td>
<td>0.211±0.014 (1.01)</td>
<td>3.68±0.277 (1.03)</td>
<td>4.75±0.472 (1.62)</td>
<td>0.023±0.0026 (1.53)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

\(^{a}(p < 0.05), \(^{b}(p < 0.01), \(^{c}(p < 0.005)\) and \(^{d}(p < 0.001)\) represent significant changes against control.

<table>
<thead>
<tr>
<th>(N)</th>
<th>Unit</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>nmole/mg protein</td>
</tr>
<tr>
<td>2</td>
<td>μmole of NADPH oxidised/min/mg protein</td>
</tr>
<tr>
<td>3</td>
<td>μmole of NADH oxidised/min/mg protein</td>
</tr>
<tr>
<td>4</td>
<td>μmole CDNB-GSH conjugate formed/min/mg protein</td>
</tr>
<tr>
<td>5</td>
<td>μmole of DCPIP reduced/min/mg protein</td>
</tr>
</tbody>
</table>

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 22. Effect of two doses of *Clerodendrum inerme* leaf extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Clerodendrum*: 50 mg/kg body wt./day and Dose II of *Clerodendrum*: 100 mg/kg body wt./day. 

a (P < 0.05), b (P < 0.01), c (P < 0.005) and d (P < 0.001) indicate significant changes against control.

Treatment duration: 14 days.
Table 33. Modulatory influence of two different doses of *Clerodendrum inerme* leaf extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatment</th>
<th>GSH (\text{\text{\textregistered}})</th>
<th>GPX (\text{\text{\textregistered}})</th>
<th>GR (\text{\text{\textregistered}})</th>
<th>SOD (\text{\text{\textregistered}})</th>
<th>CAT (\text{\text{\textregistered}})</th>
<th>LPO (\text{\text{\textregistered}})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>19.8±3.54 (1.00)</td>
<td>60.3±3.71 (1.00)</td>
<td>31.9±0.758 (1.00)</td>
<td>2.88±0.117 (1.00)</td>
<td>67.0±4.94 (1.00)</td>
<td>0.912±0.085 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Clerodendrum</em> (50 mg/kg body wt.)</td>
<td>42.0±4.57^c (2.12)</td>
<td>79.4±4.69^b (1.32)</td>
<td>40.4±3.882^b (1.27)</td>
<td>3.80±0.396^c (1.32)</td>
<td>78.4±5.44^b (1.17)</td>
<td>0.820±0.062 (0.90)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Clerodendrum</em> (100 mg/kg body wt.)</td>
<td>34.4±4.65^d (1.74)</td>
<td>110.1±6.09^b (1.82)</td>
<td>43.3±2.588^b (1.36)</td>
<td>5.50±0.539^c (1.91)</td>
<td>89.0±7.93^b (1.33)</td>
<td>0.665±0.043^c (0.73)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

^b (p < 0.01), ^c (p < 0.005) and ^d (P < 0.001) represent significant changes against control.

1nmole GSH/g tissue, 2nmole of NADPH consumed/min/mg protein, 3specific activity expressed as μmole/mg protein, 4μmole H$_2$O$_2$ consumed/min/mg protein and 5nmole malondialdehyde formed/mg protein.


Treatment duration: 14 days
Figure 23. Effect of two doses of *Clerodendrum inerme* leaf extract on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Clerodendrum*: 50 mg/kg body wt./day and Dose II of *Clerodendrum*: 100 mg/kg body wt./day.

b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control.

Treatment duration: 14 days.
Table 34. Modulatory influence of two different doses of *Clerodendrum inerme* leaf extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST 1</th>
<th>DTD 2</th>
<th>SOD 3</th>
<th>CAT 4</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.804±0.067 (1.00)</td>
<td>0.085±0.009 (1.00)</td>
<td>0.014±0.0004 (1.00)</td>
<td>5.76±0.465 (1.00)</td>
<td>23.2±2.14 (1.00)</td>
<td>3.78±0.200 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td>Clerodendrum (50 mg/kg body wt.)</td>
<td>0.860±0.085 (1.07)</td>
<td>0.109±0.012 d (1.28)</td>
<td>0.014±0.0005 (1.00)</td>
<td>6.13±0.619 (1.06)</td>
<td>32.9±3.64 c (1.42)</td>
<td>4.04±0.221 b (1.07)</td>
</tr>
<tr>
<td>Gr III</td>
<td>Clerodendrum (100 mg/kg body wt.)</td>
<td>0.90±0.058 a (1.12)</td>
<td>0.134±0.014 d (1.58)</td>
<td>0.014±0.0011 (1.00)</td>
<td>6.21±0.530 (1.08)</td>
<td>32.1±3.45 c (1.38)</td>
<td>4.28±0.275 c (1.13)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to lung in liver of control mice).

a (p < 0.05), b (p < 0.01) c (p < 0.005) and d (p < 0.001) represent significant changes against control.

1 μmole CDNB-GSH conjugate formed/min/mg protein, 2 μmole DCPIP reduced/min/mg protein
3 specific activity expressed as μmole/mg protein and 4 μmole H 2 O 2 consumed/min/mg protein.


Treatment duration: 14 days
Table 35. Modulatory influence of two different doses of *Clerodendrum inerme* leaf extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST  (µmole/min/mg protein)</th>
<th>DTD  (µmole/min/mg protein)</th>
<th>SOD  (µmole/min/mg protein)</th>
<th>CAT  (µmole/min/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>1.12±0.095 (1.00)</td>
<td>0.294±0.016 (1.00)</td>
<td>0.035±0.0035 (1.00)</td>
<td>4.04±0.300 (1.00)</td>
<td>110.3±10.9 (1.00)</td>
<td>4.07±0.239 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Clerodendrum</em> (50 mg/kg body wt.)</td>
<td>1.28±0.122 (1.14)</td>
<td>0.318±0.023 (1.08)</td>
<td>0.042±0.0041 (1.20)</td>
<td>4.10±0.572 (1.02)</td>
<td>142.0±14.5 (1.29)</td>
<td>4.08±0.290 (1.00)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Clerodendrum</em> (100 mg/kg body wt.)</td>
<td>1.19±0.095 (1.06)</td>
<td>0.345±0.030 (1.17)</td>
<td>0.044±0.0030 (1.26)</td>
<td>4.65±0.670 (1.15)</td>
<td>153.4±12.6 (1.39)</td>
<td>4.2±0.237 (1.03)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to kidney of control mice).

• (p < 0.05), b (p < 0.01), c (p < 0.005) and d (p < 0.001) represent significant changes against control.

(1) µmole CDNB-GSH conjugate formed/min/mg protein, (2) µmole DCPIP reduced/min/mg protein, (3) specific activity expressed as µmole/mg protein and (4) µmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 36. Modulatory influence of two different doses of *Clerodendrum inerme* leaf extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.174±0.014 (1.00)</td>
<td>0.272±0.032 (1.00)</td>
<td>0.017±0.0018 (1.00)</td>
<td>6.00±0.510 (1.00)</td>
<td>ND</td>
<td>4.45±0.405 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td>Clerodendrum (50 mg/kg body wt.)</td>
<td>0.158±0.019 (0.91)</td>
<td>0.275±0.035 (1.01)</td>
<td>0.019±0.0008 (1.12)</td>
<td>7.15±0.510 b</td>
<td>ND</td>
<td>4.51±0.246 (1.01)</td>
</tr>
<tr>
<td>Gr III</td>
<td>Clerodendrum (100 mg/kg body wt.)</td>
<td>0.180±0.023 (1.03)</td>
<td>0.280±0.032 (1.03)</td>
<td>0.017±0.0013 (1.00)</td>
<td>6.90±0.620 (1.15)</td>
<td>ND</td>
<td>5.05±0.392 a (1.14)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

① (p < 0.05) and ② (p < 0.01) represent significant changes against control.

① μmole CDNB-GSH conjugate formed/min/mg protein, ② μmole DCPIP reduced/min/mg protein ③ specific activity expressed as μmole/mg protein and ④ μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Figure 24. Effect of two doses of *Clerodendrum inerme* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation.

Co: control, Dose I of *Clerodendrum*: 50 mg/kg body wt/day and Dose II of *Clerodendrum*: 100 mg/kg body wt/day.

a(P < 0.05), b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control.

Catalase activity in forestomach was not detectable.

Treatment duration: 14 days.
4.7 Studies on *Lawsonia alba*

The results of present investigation are depicted in Tables 37-42 and Figures 25-28. The body weight and body weight gain of mice, treated with *Lawsonia* root extract did not show any significant alteration as compared to those in control mice.

4.7.1 Hepatic studies

The protein levels estimated in microsomal and cytosolic fractions of liver, showed significant increase at higher dose level of treatment (1.25 and 1.35 folds, respectively) while lower dose level of treatment did not show any significant change as compared to control values. The relative weight of liver presented a significant elevation only at lower dose level of treatment over its control value (Table 37).

4.7.1.1 Cytochrome P450 system

The major components of cytochrome P450 system measured, were cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase. *Lawsonia* treatment presented a significant decrease in the levels of Cyt P450 and Cyt b5 in a dose response related manner, while Cyt b5 showed a decrease only at lower dose level of treatment. Cyt P450 level was reduced by 15% (P < 0.05) and 17% (P < 0.005) at lower and higher dose levels, respectively, as compared to the control values. Cyt b5 level was reduced by 19% (P < 0.005) at higher dose level of treatment against its control value.

Specific activities of Cyt P450 reductase and Cyt b5 reductase remained almost unaffected at higher dose level of treatment whereas lower dose of treatment resulted into a significant elevation of 1.19 folds (P < 0.01) in the activity Cyt b5 reductase. In case of Cyt P450 reductase an increase of 1.11 folds was seen in Group II which was not significant (Table 38, Figure 26).
4.7.1.2 Phase II enzymes

The cytosolic fraction was used for determining the activities of phase II enzymes, glutathione S-transferase and DT-diaphorase. GST and DTD showed significant elevation in their specific activities at lower dose level of treatment by 1.11 folds (P < 0.05) and 1.13 folds (P < 0.05) respectively, as compared to their control values. The increase in the activities of GST and DTD, at higher dose level of treatment (9% and 8%, respectively) was not found statistically significant when compared to the value in the untreated groups (Table 38, Figure 26).

4.7.1.3 Antioxidative parameters

Administration of *Lawsonia* resulted in a significant as well as dose dependent enhancement in the level of reduced glutathione measured as acid soluble sulphhydryl group (-SH). The -SH level was elevated by 1.40 folds (P < 0.005) in Group II and 2.52 folds (P < 0.005) in Group III as compared to that in control group. The antioxidant enzymes measured in cytosolic fraction of liver were glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase. The significant as well as maximum induction in the specific activities of these enzymes were seen at lower dose level of treatment (1.30, 1.10, 1.50 and 1.24 folds respectively). At higher dose level of treatment, the activities of GPX, SOD and catalase were increased by 1.23, 1.45 and 1.15 folds (P < 0.005) respectively (Table 39, Figure 27).

4.7.1.4 Lipid peroxidation and lactate dehydrogenase

MDA formation was taken as an indicator of lipid peroxidation which showed a significant as well as dose dependent inhibition against the value in the untreated group. MDA formation was inhibited by 32% (P < 0.005) in Group II and 40% (P < 0.005) in Group III (Table 39). The decrease in the specific activity of lactate dehydrogenase in *Lawsonia* treated groups of mice was not significant as compared to control value (Table 37).
4.7.2 Extrahepatic studies

All the extrahepatic organs examined in the present study (lung, kidney and forestomach) did not show any significant alterations in their relative weights following *Lawsonia* treatment as compared to their control values, except lung, in which a significant increase ($P < 0.05$) was seen at higher dose level of treatment.

In kidney and forestomach, the protein levels were found almost unaltered in both extract treated groups. In lung, the protein level was significantly increased in both *Lawsonia* treated groups i.e. Group II and Group III by 25% and 35% respectively as compared to the control value (Tables 40-42).

4.7.2.1 Glutathione S-transferase

*Lawsonia* root extract significantly induced the GST activity in all the three extrahepatic organs (lung, kidney and forestomach) at both the dose levels of treatment over that of their controls, except in forestomach, in which higher dose treated groups did not cause any alteration. At lower dose level of treatment, the specific activity of GST was elevated by 1.31 ($P < 0.005$), 1.15 ($P < 0.05$) and 1.15 ($P < 0.05$) folds in lung, kidney and forestomach respectively whereas in lung and kidney of mice treated with the higher dose the activity of GST was 1.15 ($P < 0.05$) and 1.19 ($P < 0.01$) folds respectively (Tables 40-42).

4.7.2.2 DT-diaphorase

Administration of *Lawsonia* root extract resulted in a significant induction in the specific activity of DT-diaphorase in lung, kidney and forestomach except in the kidney of higher dose treated mice. The lower dose of extract (50 mg/kg body wt) increased the activity of DTD 1.18 ($P < 0.005$), 1.24 ($P < 0.01$) and 1.32 ($P < 0.001$) folds in lung, kidney and forestomach respectively. In higher dose treated group of mice DTD activity was elevated by 1.18 folds in lung and 1.16 folds in forestomach (Tables 40-42, Figure 28).
4.7.2.3 Superoxide dismutase

The increase in specific activity of superoxide dismutase following *Lawsonia* treatment, was not found significant in kidney and forestomach as compared to values in the untreated mice. In lung, SOD activity was enhanced by 1.36 folds ($P < 0.005$) at higher dose level of treatment whereas in lower dose treated group it was comparable to the control value (Tables 40-42, Figure 28).

4.7.2.4 Catalase

The catalase activity detectable only in lung and kidney under our present assay conditions, showed a dose dependent and significant increase in lung following extract treatment (1.24 folds in Group II and 1.51 folds in Group III). The specific activity of catalase in kidney was elevated at both dose levels of treatment though it was found significant only in lower dose treated group of mice (Group II; 1.31 folds, $P < 0.05$), (Tables 40-41, Figure 28).
Table 37. Modulatory influence of two different doses of *Lawsonia alba* root extract on body weight gain and toxicity related parameters.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt. x 100/ Final body wt.</th>
<th>LDH (μmole/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td>Microsome</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>25.3±1.035 (1.00)</td>
<td>26.0±1.852 (1.00)</td>
<td>4.56±0.147 (1.00)</td>
<td>2.89±0.354 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em> (50 mg/kg body wt.)</td>
<td>25.5±0.926 (1.01)</td>
<td>27.4±0.976 (1.05)</td>
<td>4.90±0.299a (1.07)</td>
<td>2.67±0.170 (0.92)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em> (100 mg/kg body wt.)</td>
<td>25.3±1.035 (1.00)</td>
<td>26.6±1.512 (1.02)</td>
<td>4.36±0.167 (0.96)</td>
<td>2.82±0.326 (0.98)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

a *(p < 0.05)* and b *(p < 0.01)* represent significant changes against control.

①μmole/mg protein.

Abbreviations- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 25. Effect of two doses of *Lawsonia alba* root extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Lawsonia*: 50 mg/kg body wt./day and Dose II of *Lawsonia*: 100 mg/kg body wt./day.

a(P < 0.05) and b(P < 0.01) indicate significant changes against control.

Treatment duration: 14 days.
Table 38. Modulatory influence of two different doses of *Lawsonia alba* root extract on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 (1)</th>
<th>Cyt b5 (2)</th>
<th>Cyt P450 R (3)</th>
<th>Cyt b5 R (4)</th>
<th>GST (5)</th>
<th>DTD (6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>0.986±0.063 (1.00)</td>
<td>0.290±0.022 (1.00)</td>
<td>0.224±0.024 (1.00)</td>
<td>4.32±0.339 (1.00)</td>
<td>5.53±0.433 (1.00)</td>
<td>0.024±0.0016 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em> (50 mg/kg body wt.)</td>
<td>0.839±0.076a (0.85)</td>
<td>0.269±0.017 (0.93)</td>
<td>0.248±0.022 (1.11)</td>
<td>5.14±0.358b (1.19)</td>
<td>6.13±0.292a (1.11)</td>
<td>0.027±0.0021a (1.13)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em> (100 mg/kg body wt.)</td>
<td>0.822±0.040c (0.83)</td>
<td>0.235±0.023c (0.81)</td>
<td>0.244±0.021 (1.09)</td>
<td>4.42±0.286 (1.02)</td>
<td>6.03±0.646 (1.09)</td>
<td>0.026±0.0019 (1.08)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

a (p < 0.05), b (p < 0.01) and c (p < 0.005) represent significant changes against control.

1 nmole/mg protein, 2 μmole of NADPH oxidised/min/mg protein, 3 μmole of NADH oxidised/min/mg protein, 4 μmole CDNB-GSH conjugate formed/min/mg protein and 5 μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 26. Effect of two doses of *Lawsonia alba* root extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-Cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Lawsonia*: 50 mg/kg body wt./day and Dose II of *Lawsonia*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 39. Modulatory influence of two different doses of *Lawsonia alba* root extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>GSH 1</th>
<th>GPX 2</th>
<th>GR 3</th>
<th>SOD 4</th>
<th>CAT 5</th>
<th>LPO 6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>13.9±1.21 (1.00)</td>
<td>65.1±3.12 (1.00)</td>
<td>50.3±3.06 (1.00)</td>
<td>6.77±0.525 (1.00)</td>
<td>67.5±2.98 (1.00)</td>
<td>1.032±0.137 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em> (50 mg/kg body wt.)</td>
<td>19.5±2.85c (1.40)</td>
<td>84.4±3.16c (1.30)</td>
<td>55.4±2.85a (1.10)</td>
<td>10.18±0.976c (1.50)</td>
<td>83.5±7.85c (1.24)</td>
<td>0.707±0.064c (0.68)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em> (100 mg/kg body wt.)</td>
<td>35.1±3.06c (2.52)</td>
<td>80.3±4.29c (1.23)</td>
<td>50.0±2.99 (0.99)</td>
<td>1.79±0.976c (1.45)</td>
<td>77.5±4.84c (1.15)</td>
<td>0.624±0.065c (0.60)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

^a (p < 0.05) and ^c (p < 0.005) represent significant changes against control.

®nmole GSH/g tissue, ®nmole of NADPH consumed/min/mg protein, ®specific activity expressed as µmole/mg protein, ®µmole H₂O₂ consumed/min/mg protein and ®nmole malondialdehyde formed/mg protein.


Treatment duration: 14 days
Figure 27. Effect of two doses of *Lawsonia alba* root extract on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice.

Error bars represent standard deviation.
Co: control, Dose I of *Lawsonia*: 50 mg/kg body wt./day and Dose II of *Lawsonia*: 100 mg/kg body wt./day.
a(P < 0.05) and c(P < 0.005) indicate significant changes against control.
Treatment duration: 14 days.
Table 40. Modulatory influence of two different doses of *Lawsonia alba* root extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST ((\mu)mole/min/mg protein)</th>
<th>DTD ((\mu)mole/min/mg protein)</th>
<th>SOD ((\mu)mole/min/mg protein)</th>
<th>CAT ((\mu)mole/min/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>0.585±0.055 (1.00)</td>
<td>0.362±0.026 (1.00)</td>
<td>0.011±0.0005 (1.00)</td>
<td>3.86±0.343 (1.00)</td>
<td>45.7±3.84 (1.00)</td>
<td>5.84±0.390 (1.00)</td>
</tr>
<tr>
<td></td>
<td>(only vehicle-d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em> (50 mg/kg body wt.)</td>
<td>0.609±0.035 (1.04)</td>
<td>0.475±0.054 (1.31)</td>
<td>0.013±0.0012 (1.18)</td>
<td>4.00±0.369 (1.04)</td>
<td>56.8±7.29 (1.24)</td>
<td>4.40±0.421 (0.75)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em> (100 mg/kg body wt.)</td>
<td>0.673±0.056 (1.15)</td>
<td>0.417±0.032 (1.15)</td>
<td>0.013±0.0009 (1.18)</td>
<td>5.23±0.459 (1.36)</td>
<td>69.0±2.89 (1.51)</td>
<td>3.78±0.285 (0.65)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to activity in lung of control mice).

\(a (p < 0.05)\), \(b (p < 0.01)\), \(c (p < 0.005)\) and \(d (p < 0.001)\) represent significant changes against control.

\(\text{GST: glutathione S-transferase, DTD: DT-diaphorase, SOD: superoxide dismutase and CAT: catalase.}\)

Treatment duration: 14 days
Table 41. Modulatory influence of two different doses of *Lawsonia alba* root extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST 1 (μmol CDNB-GSH conjugate formed/min/mg protein)</th>
<th>DTD 2 (μmol DCPIP reduced/min/mg protein)</th>
<th>SOD 3 (%)</th>
<th>CAT 4 (%)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>1.23±0.127 (1.00)</td>
<td>0.306±0.027 (1.00)</td>
<td>0.027±0.00034 (1.00)</td>
<td>3.82±0.489 (1.00)</td>
<td>102.7±18.54 (1.00)</td>
<td>6.77±0.582 (1.00)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(only vehicle-d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em></td>
<td>1.25±0.092 (1.02)</td>
<td>0.353±0.019(^a)</td>
<td>0.033±0.0023(^b)</td>
<td>4.00±0.456 (1.05)</td>
<td>134.2±6.89(^a)</td>
<td>6.38±0.493 (0.94)</td>
</tr>
<tr>
<td></td>
<td>(50 mg/kg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>body wt.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em></td>
<td>1.26±0.074 (1.02)</td>
<td>0.364±0.026(^b)</td>
<td>0.028±0.0025</td>
<td>4.39±0.431 (1.15)</td>
<td>13.5±8.15 (1.10)</td>
<td>6.56±0.246 (0.97)</td>
</tr>
<tr>
<td></td>
<td>(100 mg/kg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>body wt.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to activity in kidney of control mice).

\(^a\) (p < 0.05) and \(^b\) (p < 0.01) represent significant changes against control.

1μmol CDNB-GSH conjugate formed/min/mg protein, 2μmol DCPIP reduced/min/mg protein 3Specific activity expressed as μmol/mg protein and 4μmol H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 42. Modulatory influence of two different doses of *Lawsonia alba* root extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>0.145±0.023 (1.00)</td>
<td>0.215±0.017 (1.00)</td>
<td>0.019±0.0011 (1.00)</td>
<td>5.76±0.465 (1.00)</td>
<td>ND</td>
<td>4.51±0.194 (1.00)</td>
</tr>
<tr>
<td></td>
<td>(only vehicle- d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Lawsonia</em> (50 mg/kg body wt.)</td>
<td>0.149±0.013 (1.03)</td>
<td>0.239±0.016 a (1.11)</td>
<td>0.025±0.0021 d (1.32)</td>
<td>6.21±0.819 (1.08)</td>
<td>ND</td>
<td>4.57±0.452 (1.01)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Lawsonia</em> (100 mg/kg body wt.)</td>
<td>0.157±0.013 (1.08)</td>
<td>0.215±0.013 (1.00)</td>
<td>0.022±0.0015 c (1.16)</td>
<td>6.13±0.730 (1.06)</td>
<td>ND</td>
<td>4.43±0.458 (0.98)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

a (p < 0.05), c (p < 0.005) and d (p < 0.001) represent significant changes against control.

①μmole CDNB-GSH conjugate formed/min/mg protein, ②μmole DCPIP reduced/min/mg protein
③specific activity expressed as μmole/mg protein and ④μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Figure 28. Effect of two doses of *Lawsonia alba* root extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation.

Co: control, Dose I of *Lawsonia*: 50 mg/kg body wt/day and Dose II of *Lawsonia*: 100 mg/kg body wt/day.

a(P < 0.05), b(P < 0.01), c(P < 0.005) and d(P < 0.001) indicate significant changes against control.

Catalase activity in forestomach was not detectable.

Treatment duration: 14 days.
4.8 Studies on *Prosopis juliflora*

The results of the present investigation in liver, lung, kidney and forestomach of mice following treatment with hydroalcoholic extract of leaves of *Prosopis* are illustrated in Tables 43-48 and Figures 29-32.

The body weight and body weight gain of mice treated with *Prosopis* remained unaltered as in untreated group of mice.

4.8.1 Hepatic studies

The relative weight of liver was increased in mice receiving extract but was significant only at higher dose level of treatment. (Group III; 1.22 folds, \( P < 0.01 \)). The protein levels in microsomal and cytosolic fractions did not show any significant decrease at both the dose levels of treatment as compared to respective control values (Table 43).

4.8.1.1 Cytochrome P450 system

The major components of cytochrome P450 system analyzed in the microsomal fraction of liver of mice were cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase. The level of Cyt P450 showed a significant decrease at lower dose level of treatment \((P < 0.01)\); in contrast the level of Cyt b5 showed a significant elevation at higher dose level of treatment \((P < 0.05)\) against control. The specific activity of Cyt P450 reductase increased in a dose dependent manner and were 1.16 folds \((P < 0.05)\) in Group II and 1.21 folds \((P < 0.05)\) in Group III as compared to its control values. *Prosopis* administration did not affect the activity of Cyt b5 reductase (Table 44, Figure 30).

4.8.1.2 Phase II enzymes

For studying the effect of *Prosopis* leaf extract on phase II enzymes, the specific activities of glutathione S-transferase and DT-diaphorase were measured. The activity of GST was increased in a dose dependent manner and was 1.22 \((P < 0.05)\) and 1.52 \((P < 0.005)\) folds in lower and higher dose treated groups respectively. DTD activity was
increased by 1.16 folds \((P < 0.05)\) at lower dose level of treatment as compared to that in control (Table 44, Figure 30).

4.8.1.3 Antioxidative parameters

The antioxidative parameters measured were reduced glutathione, glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase. The content of reduced glutathione measured as acid soluble sulphydryl group in liver homogenate showed significant decrease \((P < 0.05)\) at higher dose level of treatment as compared to that in untreated group. The specific activity of GPX was elevated by 1.27 folds \((P < 0.005)\) in higher dose treated group of mice. GR and SOD did not show any significant alteration following treatment with test substance. The specific activity of catalase was enhanced in a dose dependent manner by 1.11 \((P < 0.05)\) folds in Group II and 1.47 \((P < 0.005)\) folds in Group III relative to its control group (Table 45, Figure 31).

4.8.1.4 Lipid peroxidation and lactate dehydrogenation

The administration of the Prosopis leaf extract resulted in a significant as well as dose dependent inhibition of lipid peroxidation, which was reduced by 17\% \((P < 0.05)\) and 31\% \((P < 0.005)\) at lower and higher dose levels of treatment, respectively (Table 45). The specific activity of lactate dehydrogenase was significantly reduced by 13\% \((P < 0.05)\) at lower dose level of treatment as compared to its control value (Table 43).

4.8.2 Extrahepatic studies

The mice treated with Prosopis leaf extract did not show any significant alterations in the relative weights of lung, kidney and forestomach as compared to their controls. The protein levels in lung and forestomach did not show any significant deviation from their control values whereas in kidney, it was significantly reduced by 13\% and 14\% in lower and higher dose treated groups respectively (Tables 46-48).
4.8.2.1 Glutathione S-transferase

A distinct dose dependent induction in the activity of glutathione S-transferase was evident in lung, kidney as well as forestomach, following administration of *Prosopis* leaf extract. The increase in the activity of GST in lung was significant in higher dose treated group of mice and it was 1.29 folds (P < 0.001) as compared to that in control. In kidney and forestomach, at lower dose level of treatment GST activity was elevated by 1.29 (P < 0.005) and 1.39 (P < 0.01) folds respectively. In higher dose treated group, the GST activity increased by 1.65 folds (P < 0.005) in kidney and 1.49 folds (P < 0.001) in forestomach as compared to their respective control values (Tables 46-48, Figure 32).

4.8.2.2 DT-diaphorase

The activity of DTD was significantly elevated in lung, kidney and forestomach of mice following treatment with *Prosopis*. The enhancement in the activity of DTD in lung and kidney was dose dependent and was 1.09 (P < 0.05) and 1.13 (P < 0.001) folds in lower dose treated group; and 1.14 (P < 0.01) and 1.42 (P < 0.005) folds in higher dose treated group respectively, as compared to their control values. In case of forestomach, the induced level of DTD was 1.17 folds (P < 0.005) in both the extract treated groups in relation to the control values (Tables 46-48, Figure 32).

4.8.2.3 Superoxide dismutase

Activity of superoxide dismutase remained unaffected in the lung of mice following treatment with *Prosopis*. Kidney and forestomach showed significant elevation in SOD activity in both the extract treated groups as compared to their respective control values. In kidney, the activity of SOD was increased by 2.02 folds (P < 0.005) in Group II and 1.98 folds (P < 0.01) in Group III, and in forestomach by 1.71 folds (P < 0.005) in Group II and 2.02 folds (P < 0.005) in Group III (Tables 46-48, Figure 32).

4.8.2.4 Catalase

Catalase activity, detectable only in lung and kidney under our assay conditions, showed significant as well as dose dependent enhancement following treatment with
Prosopis. At lower dose level of treatment, the catalase activity was increased by 1.30 folds ($P < 0.005$) in lung and 1.36 folds ($P < 0.005$) in kidney; and at higher dose level by 1.38 folds ($P < 0.005$) and 1.66 folds ($P < 0.005$) respectively, as compared to their respective control values (Tables 46-47, Figure 32).
Table 43. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on body weight gain and toxicity related parameters.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt.x100/ Final body wt.</th>
<th>LDH (\text{\textmu}mol/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>Final</td>
<td></td>
<td>Microsome</td>
</tr>
<tr>
<td>Gr I</td>
<td>Control (only vehicle- d.w.)</td>
<td>27.1±0.354 (1.00)</td>
<td>29.3±0.463 (1.00)</td>
<td>5.06±0.476 (1.00)</td>
<td>2.02±0.220 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em> (50 mg/kg body wt.)</td>
<td>27.9±0.354 (0.99)</td>
<td>29.3±0.463 (1.00)</td>
<td>5.30±0.240 (1.05)</td>
<td>1.75±0.158(\boldsymbol{a}) (0.87)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em> (100 mg/kg body wt.)</td>
<td>27.1±0.354 (1.00)</td>
<td>29.6±1.063 (1.01)</td>
<td>6.17±0.527(\boldsymbol{b}) (1.22)</td>
<td>2.11±0.245 (1.04)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

\(\boldsymbol{a}\) (p < 0.05) and \(\boldsymbol{b}\) (p < 0.01) represent significant changes against control.

\(\text{\textmu}mol/mg\) protein.

Abbreviations- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 29. Effect of two doses of *Prosopis juliflora* leaf extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Prosopis*: 50 mg/kg body wt./day and Dose II of *Prosopis*: 100 mg/kg body wt./day. a(P < 0.05) and b(P < 0.01) indicate significant changes against control. Treatment duration: 14 days.
Table 44. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on mouse hepatic phase I and phase II drug metabolising enzyme levels.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 1</th>
<th>Cyt b5 2</th>
<th>Cyt P450 R 3</th>
<th>Cyt b5 R 4</th>
<th>GST 5</th>
<th>DTD 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.691±0.051 (1.00)</td>
<td>0.319±0.026 (1.00)</td>
<td>0.259±0.024 (1.00)</td>
<td>2.92±0.215 (1.00)</td>
<td>3.12±0.408 (1.00)</td>
<td>0.025±0.0009 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em> (50 mg/kg body wt.)</td>
<td>0.602±0.035&lt;sup&gt;b&lt;/sup&gt; (0.87)</td>
<td>0.341±0.032 (1.07)</td>
<td>0.301±0.032&lt;sup&gt;a&lt;/sup&gt; (1.16)</td>
<td>2.91±0.152 (1.00)</td>
<td>3.80±0.161&lt;sup&gt;a&lt;/sup&gt; (1.22)</td>
<td>0.029±0.0014&lt;sup&gt;c&lt;/sup&gt; (1.16)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em> (100 mg/kg body wt.)</td>
<td>0.629±0.058&lt;sup&gt;a&lt;/sup&gt; (0.91)</td>
<td>0.363±0.031&lt;sup&gt;a&lt;/sup&gt; (1.14)</td>
<td>0.314±0.032&lt;sup&gt;a&lt;/sup&gt; (1.21)</td>
<td>2.90±0.140 (0.99)</td>
<td>4.73±0.296&lt;sup&gt;c&lt;/sup&gt; (1.52)</td>
<td>0.026±0.0020 (1.04)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

<sup>a</sup>(p < 0.05), <sup>b</sup>(p < 0.01) and <sup>c</sup>(p < 0.005) represent significant changes against control.

①nmole/mg protein, ②μmole of NADPH oxidised/min/mg protein, ③μmole of NADH oxidised/min/mg protein ④μmole CDNB-GSH conjugate formed/min/mg protein and ⑤μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: cytochrome P450 reductase, Cyt b5 R: cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 30. Effect of two doses of *Prosopis juliflora* leaf extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Prosopis*: 50 mg/kg body wt./day and Dose II of *Prosopis*: 100 mg/kg body wt./day. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control. Treatment duration: 14 days
Table 45. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>GSH ((\text{GSH} / \text{g tissue}))</th>
<th>GPX ((\text{nmole of NADPH consumed/min/mg protein}))</th>
<th>GR ((\text{specific activity expressed as } \mu \text{mole/mg protein}))</th>
<th>SOD ((\text{nmole } H_2O_2 \text{ consumed/min/mg protein}))</th>
<th>CAT ((\text{nmole malondialdehyde formed/mg protein}))</th>
<th>LPO ((\text{nmole } \text{malondialdehyde formed/mg protein}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>30.8±3.67</td>
<td>58.4±5.51</td>
<td>28.8±2.94</td>
<td>6.30±0.512</td>
<td>82.7±5.43</td>
<td>0.830±0.080</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(1.00)</td>
<td>(1.00)</td>
<td>(1.00)</td>
<td>(1.00)</td>
<td>(1.00)</td>
<td>(1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em></td>
<td>32.2±4.46</td>
<td>59.6±5.1</td>
<td>31.4±1.94</td>
<td>6.50±0.793</td>
<td>91.7±5.80</td>
<td>0.692±0.618</td>
</tr>
<tr>
<td></td>
<td>(50 mg/kg body wt.)</td>
<td></td>
<td>(1.04)</td>
<td>(1.02)</td>
<td>(1.09)</td>
<td>(1.03)</td>
<td>(1.11)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em></td>
<td>25.1±4.48</td>
<td>74.1±4.34</td>
<td>31.3±1.67</td>
<td>6.09±0.626</td>
<td>121.6±6.94</td>
<td>0.575±0.049</td>
</tr>
<tr>
<td></td>
<td>(100 mg/kg body wt.)</td>
<td></td>
<td>(0.82)</td>
<td>(1.27)</td>
<td>(1.09)</td>
<td>(0.97)</td>
<td>(1.47)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

\(^{a}(p < 0.05)\) and \(^{c}(p < 0.005)\) represent significant changes against control.

\(\text{GSH: reduced glutathione, GPX: glutathione peroxidase, GR: glutathione reductase, SOD: superoxide dismutase, CAT: catalase and LPO: lipid peroxidation.}\)


Treatment duration: 14 days
Figure 31. Effect of two doses of *Prosopis juliflora* leaf extract on the level of reduced glutathione (GSH), on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Prosopis*: 50mg/kg body wt./day and Dose II of *Prosopis*: 100mg/kg body wt./day.

a(P < 0.05) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 46. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on detoxifying and antioxidant enzyme profiles in lung of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of lung (%)</th>
<th>GST ①</th>
<th>DTD ②</th>
<th>SOD ③</th>
<th>CAT ④</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.801±0.053 (1.00)</td>
<td>0.249±0.019 (1.00)</td>
<td>0.022±0.0014 (1.00)</td>
<td>6.50±0.473 (1.00)</td>
<td>16.65±1.54 (1.00)</td>
<td>4.28±0.322 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em> (50 mg/kg body wt.)</td>
<td>0.581±0.039 (1.03)</td>
<td>0.026±0.023 (1.05)</td>
<td>0.024±0.0011a (1.09)</td>
<td>5.87±0.601 (0.90)</td>
<td>21.68±2.27c (1.30)</td>
<td>4.34±0.295 (1.01)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em> (100 mg/kg body wt.)</td>
<td>0.825±0.039 (1.05)</td>
<td>0.322±0.038d (1.29)</td>
<td>0.025±0.0021b (1.14)</td>
<td>4.08±0.369b (0.63)</td>
<td>22.91±1.73c (1.38)</td>
<td>4.45±0.334 (1.04)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in lung of mice receiving test substance to lung in liver of control mice).

a (p < 0.05), b (p < 0.01), c (p < 0.005) and d (p < 0.001) represent significant changes against control.

① μmole CDNB-GSH conjugate formed/min/mg protein, ② μmole DCPIP reduced/min/mg protein ③ specific activity expressed as μmole/mg protein and ④ μmole H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 47. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on detoxifying and antioxidant enzyme profiles in kidney of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of kidney (%)</th>
<th>GST (μmol CDNB-GSH conjugate formed/min/mg protein)</th>
<th>DTD (μmol DCPIP reduced/min/mg protein)</th>
<th>SOD (% of kidney of control)</th>
<th>CAT (specific activity expressed as μmol H₂O₂ consumed/min/mg protein)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>1.46±0.072 (1.00)</td>
<td>0.368±0.016 (1.00)</td>
<td>0.024±0.0004 (1.00)</td>
<td>3.68±0.573 (1.00)</td>
<td>83.4±6.15 (1.00)</td>
<td>5.65±0.256 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em> (50 mg/kg body wt.)</td>
<td>1.47±0.148 (1.01)</td>
<td>0.475±0.036c (1.29)</td>
<td>0.027±0.0013d (1.13)</td>
<td>7.47±0.767c (2.02)</td>
<td>113.4±5.61c (1.36)</td>
<td>4.92±0.238c (0.87)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em> (100 mg/kg body wt.)</td>
<td>1.42±0.123 (0.97)</td>
<td>0.605±0.042c (1.65)</td>
<td>0.034±0.0026c (1.42)</td>
<td>7.27±0.923b (1.98)</td>
<td>138.6±7.73c (1.66)</td>
<td>4.84±0.370c (0.86)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in kidney of mice receiving test substance to kidney of control mice).

b (p < 0.01), c (p < 0.005) and d (p < 0.001) represent significant changes against control.

1 μmol CDNB-GSH conjugate formed/min/mg protein, 2 μmol DCPIP reduced/min/mg protein 3 specific activity expressed as μmol/mg protein and 4 μmol H₂O₂ consumed/min/mg protein.


Treatment duration: 14 days
Table 48. Modulatory influence of two different doses of *Prosopis juliflora* leaf extract on detoxifying and antioxidant enzyme profiles in forestomach of mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Relative wt. of forestomach (%)</th>
<th>GST (μmol)</th>
<th>DTD (μmol)</th>
<th>SOD (Units)</th>
<th>CAT (Units)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.218±0.016 (1.00)</td>
<td>0.181±0.022 (1.00)</td>
<td>0.029±0.0018 (1.00)</td>
<td>4.17±0.523 (1.00)</td>
<td>ND</td>
<td>3.78±0.337 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Prosopis</em> (50 mg/kg body wt.)</td>
<td>0.229±0.025 (1.05)</td>
<td>0.251±0.011 (1.39)</td>
<td>0.034±0.0031 (1.17)</td>
<td>7.13±0.867 (1.71)</td>
<td>ND</td>
<td>3.87±0.131 (1.02)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Prosopis</em> (100 mg/kg body wt.)</td>
<td>0.208±0.009 (0.95)</td>
<td>0.269±0.021 (1.49)</td>
<td>0.034±0.0026 (1.17)</td>
<td>8.41±0.383 (2.02)</td>
<td>ND</td>
<td>3.84±0.380 (1.02)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in forestomach of mice receiving test substance to activity in forestomach of control mice).

\( ^b (p < 0.01), ^c (p < 0.005) \) and \( ^d (p < 0.001) \) represent significant changes against control.

\( ^1 \mu \text{mole CDBN-GSH conjugate formed/min/mg protein, } ^2 \mu \text{mole DCPIP reduced/min/mg protein} \)
\( ^3 \text{specific activity expressed as } \mu \text{mole/mg protein and } ^4 \mu \text{mole H}_2\text{O}_2 \text{ consumed/min/mg protein.} \)


Treatment duration: 14 days
Figure 32. Effect of two doses of *Prosopis juliflora* leaf extract on the specific activities of glutathione S-transferase (GST), DT-diaphorase (DTD), superoxide dismutase (SOD) and catalase (CAT) in lung, kidney and forestomach of mice. Error bars represent standard deviation.

Co: control, Dose I of *Prosopis*: 50 mg/kg body wt/day and Dose II of *Prosopis*: 100 mg/kg body wt/day.

a(p < 0.05), b(p < 0.01), c(p < 0.005) and d(p < 0.001) indicate significant changes against control.

Catalase activity in forestomach was not detectable.

Treatment duration: 14 days.
4.9 Studies on *Decalepis hamiltonii*

The treatment of mice with hydroalcoholic extract of *Decalepis* root lead to the various changes in the parameters investigated in liver (Tables 49-51 and Figures 33-35).

Mice treated with *Decalepis* did not show any significant change in body weight and body weight gain except high dose treated group of mice in which final body weight was seen significantly increased ($P < 0.05$) as compared to control.

4.9.1 Hepatic studies

The liver-somatic index in *Decalepis* treated groups did not show any significant change against that in untreated group. The protein level in microsomal fraction of liver of extract treated groups were comparable to that in control group. The cytosolic protein was significantly elevated in both groups of mice which received test substance (Group II and Group III; 1.25 and 1.21 folds respectively), (Table 49).

The microsomal fraction of liver homogenate was used for estimating the modulation of cytochrome P450 system (cytochrome P450, cytochrome b5, NADPH-cytochrome P450 reductase and NADH-cytochrome b5 reductase) and lipid peroxidation. The cytosolic fraction was used for determining the activities of phase II enzymes (GST and DTD), antioxidative parameters (GSH, GPX, GR, SOD and catalase) and lactate dehydrogenase.

4.9.1.1 Cytochrome P450 system

The content of cytochrome P450 was significantly enhanced in Group III whereas the content of cytochrome b5 was found significantly elevated in Group II. Cyt P450 level in Group III was elevated by 1.85 folds ($P < 0.005$) and Cyt b5 level in Group II was elevated by 1.22 folds ($P < 0.005$) as compared to their control values.

The specific activity of NADPH-cytochrome P450 reductase was significantly enhanced at both dose levels of *Decalepis* treatment (Group II and Group III; 1.25 and 1.25 folds) against control. The increase in the specific activity of NADH-cytochrome b5 reductase in extract treated groups was not significant as compared to the values in its untreated group (Table 50, Figure 34).
Results

4.9.1.2 Phase II enzymes

For studying the effect of *Decalepis* on phase II enzymes, the specific activities of glutathione S-transferase and DT-diaphorase were measured. The extract treatment showed an increase in the activities of both enzymes in a similar pattern. The levels of induction in GST and DTD were higher in lower dose treated group (1.50 folds, \( P < 0.005 \); and 1.39 folds, \( P < 0.01 \) respectively) than that of higher dose treated group (1.39 folds, \( P < 0.005 \); and 1.29 folds \( P < 0.005 \) respectively) (Table 50, Figure 34).

4.9.1.3 Antioxidative parameters

Reduced glutathione, glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase were measured as antioxidative parameters. Reduced glutathione measured as acid soluble sulfhydryl group (-SH) in the liver of mice was significantly increased in dose dependent manner following administration of *Decalepis* [1.31 folds (\( P < 0.01 \)) in Group II and 1.80 folds \( (P < 0.005) \) in Group III]. The changes in the specific activities of GPX and GR were not significant against their control values except the higher dose treated group of GR which showed a significant increase of 1.16 folds \( (P < 0.05) \) over its control value. SOD and catalase were significantly induced by the treatment with test substance. At lower dose level of treatment the specific activities of SOD and catalase were increased by 1.19 folds \( (P < 0.05) \) and 1.57 folds \( (P < 0.005) \); and at higher dose level by 1.32 folds \( (P < 0.005) \) and 1.30 folds \( (P < 0.005) \) respectively as compared to their control values (Table 51, Figure 35).

4.9.1.4 Lipid peroxidation and lactate dehydrogenase

MDA production, a secondary product of lipid peroxidation, was measured in microsomal fraction of liver which showed a significant as well as dose dependent inhibition following treatment of *Decalepis*. Lipid peroxidation was reduced by 22\% (\( P < 0.01 \)) in Group II and 33\% (\( P < 0.005 \)) in Group III as compared to that in control group of mice (Table 51). Extract treatment also reduced the activity of lactate dehydrogenase significantly by 30\% (\( P < 0.005 \)) in higher dose treated group of mice (Table 49).
Table 49. Modulatory influence of two different doses of *Decalepis hamiltonii* root extract on body weight gain and toxicity related parameters in mouse.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Body weight (gms)</th>
<th>Liver wt x100/ Final body wt.</th>
<th>LDH ( \uparrow ) (mg/ml)</th>
<th>Protein (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial Final</td>
<td></td>
<td>Microsome Cytosol</td>
<td></td>
</tr>
<tr>
<td>Gr I</td>
<td>Control</td>
<td>19.9±1.13 22.0±1.85</td>
<td>5.03±0.415 (1.00) (1.00)</td>
<td>2.33±0.358 (1.00)</td>
<td>10.26±1.29 (1.00)</td>
</tr>
<tr>
<td></td>
<td>(only</td>
<td></td>
<td></td>
<td>6.95±1.05</td>
<td></td>
</tr>
<tr>
<td></td>
<td>vehicle-d.w.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Decalepis</em></td>
<td>19.4±1.51 21.4±1.51</td>
<td>4.74±0.432 (0.98) (0.97)</td>
<td>2.07±0.222 (0.94) (0.89)</td>
<td>9.45±1.13 (0.92)</td>
</tr>
<tr>
<td>(50 mg/kg</td>
<td>body wt.)</td>
<td></td>
<td></td>
<td></td>
<td>8.68±1.32</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Decalepis</em></td>
<td>20.9±1.07 24.6±1.90a</td>
<td>4.59±0.371 1.64±0.121c (1.05) (1.12)</td>
<td>11.60±1.35 (0.70) (1.13)</td>
<td>8.42±0.576b</td>
</tr>
<tr>
<td>(100 mg/kg</td>
<td>body wt.)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative changes in parameters assessed (i.e., levels of parameter assessed in livers of mice receiving test substance to that of control mice).

\( ^a (p < 0.05) \), \( ^b (p < 0.01) \) and \( ^c (p <0.005) \) represent significant changes against control.

\( \uparrow \) μmole/mg protein.

Abbreviations- d.w: distilled water and LDH: lactate dehydrogenase.

Treatment duration: 14 days
Figure 33. Effect of two doses of *Decalepis hamiltonii* root extract on liver-somatic index (L-S index), specific activity of lactate dehydrogenase (LDH), microsomal protein (M-protein) and cytosolic protein (C-protein) in the liver of mice. Error bars represent standard deviation. Co: control, Dose I of *Decalepis*: 50 mg/kg body wt./day and Dose II of *Decalepis*: 100 mg/kg body wt./day. a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control. Treatment duration: 14 days.
Table 50. Modulatory influence of two different doses of *Decalepis hamiltonii* on mouse hepatic phase I and phase II drug metabolizing enzyme levels.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>Cyt P450 ①</th>
<th>Cyt b5 ①</th>
<th>Cyt P450 R ②</th>
<th>Cyt b5 R ③</th>
<th>GST ④</th>
<th>DTD ⑤</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>0.338±0.043 (1.00)</td>
<td>0.386±0.066 (1.00)</td>
<td>0.229±0.015 (1.00)</td>
<td>4.24±0.323 (1.00)</td>
<td>2.44±0.269 (1.00)</td>
<td>0.034±0.0037 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Decalepis</em> (50mg/kg body wt.)</td>
<td>0.338±0.043 (1.00)</td>
<td>0.471±0.022 ⑥ (1.22)</td>
<td>0.286±0.018 ⑥ (1.25)</td>
<td>4.36±0.158 (1.03)</td>
<td>3.65±0.560 ⑥ (1.50)</td>
<td>0.047±0.0059 ⑧ (1.39)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Decalepis</em> (100 mg/kg body wt.)</td>
<td>0.626±0.080 ⑥ (1.85)</td>
<td>0.416±0.049 (1.08)</td>
<td>0.284±0.012 ⑥ (1.25)</td>
<td>4.47±0.156 (1.05)</td>
<td>3.40±0.456 ⑥ (1.39)</td>
<td>0.044±0.0023 ⑥ (1.29)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

⑥ (p < 0.01) and ⓥ (p < 0.005) represent significant changes against control.

①nmole/mg protein, ②μmole of NADPH oxidised/min/mg protein, ③μmole of NADH oxidised/min/mg protein ④μmole CDNB-GSH conjugate formed/min/mg protein and ⑤μmole of DCPIP reduced/min/mg protein.

Abbreviations- Cyt P450: cytochrome P450, Cyt b5: cytochrome b5, Cyt P450 R: NADPH-cytochrome P450 reductase, Cyt b5 R: NADH-cytochrome b5 reductase, GST: glutathione S-transferase and DTD: DT-diaphorase.

Treatment duration: 14 days
Figure 34. Effect of two doses of *Decalepis hamiltonii* root extract on the levels of cytochrome P450 (Cyt P450), cytochrome b5 (Cyt b5) and on the specific activities of NADPH-cytochrome P450 reductase (Cyt P450 R), NADH-Cytochrome b5 reductase (Cyt b5 R), glutathione S-transferase (GST) and DT-diaphorase (DTD) in the liver of mice. Error bars represent standard deviation.

Co: control, Dose I of *Decalepis*: 50 mg/kg body wt./day and Dose II of *Decalepis*: 100 mg/kg body wt./day.

b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Figure 35. Effect of two doses of *Decalepis hamiltonii* root extract on reduced glutathione content (GSH) and on the specific activities of glutathione peroxidase (GPX), glutathione reductase (GR), superoxide dismutase (SOD), catalase (CAT) and on malondialdehyde formation (LPO) in the liver mice. Error bars represent standard deviation.

Co: control, Dose I of *Decalepis*: 50 mg/kg body wt./day and Dose II of *Decalepis*: 100 mg/kg body wt./day.

a(P < 0.05), b(P < 0.01) and c(P < 0.005) indicate significant changes against control.

Treatment duration: 14 days.
Table 51. Modulatory influence of two different doses of *Decalepis hamiltonii* root extract on mouse hepatic antioxidant related parameters and lipid peroxidation.

<table>
<thead>
<tr>
<th>Groups (Gr)</th>
<th>Treatment</th>
<th>GSH ((\text{nmole/g tissue}))</th>
<th>GPX ((\text{nmole of NADPH consumed/min/mg protein}))</th>
<th>GR ((\text{specific activity expressed as (\mu)mole/mg protein}))</th>
<th>SOD ((\text{(\mu)mole H}_2\text{O}_2) consumed/min/mg protein})</th>
<th>CAT ((\text{nmole malondialdehyde formed/mg protein}))</th>
<th>LPO ((\text{nmole malondialdehyde formed/mg protein}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gr I</td>
<td>Control (only vehicle-d.w.)</td>
<td>40.0±4.70 (1.00)</td>
<td>72.8±5.11 (1.00)</td>
<td>44.4±1.95 (1.00)</td>
<td>6.72±0.271 (1.00)</td>
<td>85.2±11.28 (1.00)</td>
<td>0.840±0.126 (1.00)</td>
</tr>
<tr>
<td>Gr II</td>
<td><em>Decalepis</em> (50 mg/kg body wt.)</td>
<td>52.5±6.88(b) (1.31)</td>
<td>70.0±6.72 (1.09)</td>
<td>48.3±5.35 (1.09)</td>
<td>8.00±0.525(c) (1.19)</td>
<td>134.0±1.95(c) (1.57)</td>
<td>0.658±0.061(b) (0.78)</td>
</tr>
<tr>
<td>Gr III</td>
<td><em>Decalepis</em> (100 mg/kg body wt.)</td>
<td>72.1±7.46(c) (1.80)</td>
<td>67.4±2.70 (0.93)</td>
<td>51.6±3.58(a) (1.16)</td>
<td>8.86±0.681(c) (1.32)</td>
<td>110.5±8.08(c) (1.30)</td>
<td>0.559±0.061(c) (0.67)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD of 6-8 animals.

Values in parentheses represent relative change in parameters assessed (i.e., levels of activity in livers of mice receiving test substance to activity in liver of control mice).

\(a (p < 0.05), b (p < 0.01)\) and \(c (p < 0.005)\) represent significant changes against control.

\(\text{\(\mu\)mole GSH/g tissue}, \text{\(\mu\)mole of NADPH consumed/min/mg protein}, \text{\(\mu\)mole H}_2\text{O}_2\) consumed/min/mg protein, \text{\(\mu\)mole malondialdehyde formed/mg protein}.\)


Treatment duration: 14 days