Antioxidant Effect of Green Tea Extract In Cadmium Chloride Intoxicated Rats

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\textbf{ABSTRACT}

The present study was undertaken to examine the inhibitory effect of the green tea (Camellia sinesis) on cadmium chloride induced antioxidant activity in liver. The levels of lipid peroxidation were assessed by estimating TBARS and lipid peroxidation and the antioxidant levels were assessed by estimating the levels of GSH, SOD, CAT and GPx. Significant increases was observed in the levels of TBARS and hydroperoxide in CdCl\textsubscript{2} treated rats. These levels were significantly decreased in CdCl\textsubscript{2} and Green tea (Camellia sinesis) extract treated rats. Further enzymatic (SOD, CAT and GPx) and non-enzymatic (GSH) antioxidants were significantly decreased in CdCl\textsubscript{2} treated rats.

\textbf{Key words:} Cadmium chloride, \textit{Camellia sinesis}, Oxidative stress.

\textbf{INTRODUCTION}

Metals and metal compounds are constituents of our national environmental due to the extensive use of various metals in the modern society, over the years. Thus there is simple opportunity for exposure to toxic metals both in and outside the work place. Cadmium chloride is a well-known hepatotoxic agent. A number of evidences advocate the role of oxidative stress in cadmium induced liver toxicity. For instance, CdCl\textsubscript{2} exposure augments H\textsubscript{2}O\textsubscript{2} production by the mitochondria in hepatic cells [1]. CdCl\textsubscript{2} leads to the depletion of glutathione and inhibit the activities of antioxidant enzymes in liver tissues [2]. Metals also induce lipid peroxidation both \textit{in vitro} and \textit{in vivo} [3].
Camellia sinesis (Family-Solanacea) is an annual herb, native of India, wild or naturalized throughout the tropics of both the hemispheres. It is valued in medicines as the leaves are of use for making cigarettes and fumigating powders for the relief of asthma. The juice of this plant is believed to be a cure of hydrophobia. The leaves are boiled and used as a poultice to relieve pain. The plant contains alkaloids such as hyoscyamine (atropine), scopolamine hyoscine, etc [4]. It is a reputed drug for dog bites and respiratory alignments [5]. The present study deals with the influence of green tea extract on Cadmium chloride induced toxicity in rats by analyzing their biochemical alteration.

MATERIALS AND METHODS

Animals
Adult male albino rats of Wistar strain weighing 170-200 g were used for the study. The rats were housed in polypropylene cage and kept under standard laboratory conditions (temperature 25±2°C; natural light-dark cycle). The rats were provided with food and water ad libitum. The commercial rat feed contained 5% fat, 21% protein, 55% nitrogen free extract and 4% fibre (w/w) with adequate minerals and vitamin contents.

Chemicals
Cadmium chloride was purchased from Sigma chemical Co. (St.Louis, MO, USA). The rest of the chemicals and biochemical’s were obtained from local firms (India) and were of analytical grade.

Treatment Schedule
The animals were randomised into experimental and control groups and divided into 4 groups of six animals each. Animals in

Group-I Control rats subcutaneously treated with isotonie saline (1 mg/kg body weight/day).
Group-II The green tea was made by soaking 15 g of instant green tea powder in 1 L of boiling distilled water for 5 minutes. The solution was filtered to make 1.5% green tea extract (GTE). This solution was provided to rats as their sole source of drinking water.
Group-3 Toxicity was induced in rats by administration of cadmium chloride (1.25 mg/kg) body weight via intraperitoneal administration.
Group-4 Rats were treated with cadmium chloride (1.25 mg/kg body weight) as in group III and as in group I.

Estimations
At the end of experimental periods of 45 days, Liver were removed from all the animals and immediately transferred to ice cold containers containing 0.9% Sodium chloride (NaCl) for various estimations of biochemical parameters include such as Thiobarbituric acid reactive substance [6]; Reduced glutathione [7]; Super oxide dismutase [8]; Catalase [9] and Glutathine peroxidase [10].

Statistical analysis
All data were expressed as mean ± standard deviation of number of experiments. The statistical significance was elevated by one-way analysis of variance (ANOVA) using SPSS version 9.0.
(SPSS, Cary, NC, USA) and Duncan’s multiple range test (DMRT) obtained the individual comparisons [11]. A value of p<0.05 was considered to indicate a significant difference between groups. Values sharing a common superscript do not differ significantly with each other at p<0.05.

RESULTS

Table 1 shows the levels of TBARS in liver tissues were found to be increased in group III rats. Group IV rats showed decreases in the levels of TBARS when compared with group III rats. Group II rats there is no significant changes in the levels of TBARS. Administration of CdCl₂ caused significant decrease in GSH concentration in group III rats. Group IV rats show increased levels of GSH. Group II rats showed no significant changes in GSH levels when compared with control.

Table.1: The levels of TBARS and GSH in liver of control and experimental animals.

<table>
<thead>
<tr>
<th>SN</th>
<th>Groups</th>
<th>GSH(µg/min/mg protein)</th>
<th>TBARS(m moles/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control</td>
<td>0.41±0.03</td>
<td>0.65±0.05</td>
</tr>
<tr>
<td>2</td>
<td>Green tea extract</td>
<td>3.39±0.25</td>
<td>0.59±0.25</td>
</tr>
<tr>
<td>3</td>
<td>Cadmium chloride</td>
<td>1.20±01.2</td>
<td>3.42±0.12</td>
</tr>
<tr>
<td>4</td>
<td>Cadmium chloride+ Green tea extract</td>
<td>0.49±0.14</td>
<td>0.49±0.15</td>
</tr>
</tbody>
</table>

Values are mean ± S.D for 6 rats in each group.

Table.2: The activities of enzymatic antioxidants in liver of control and experimental animals

<table>
<thead>
<tr>
<th>SN</th>
<th>Groups</th>
<th>CAT(µ mol of H₂O₂/min/mg protein)</th>
<th>GPx(µg/min/mg protein)</th>
<th>SOD (U/mg protein)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control</td>
<td>43.12±2.18</td>
<td>5.87±0.30</td>
<td>3.87±0.15</td>
</tr>
<tr>
<td>2</td>
<td>Green tea extract</td>
<td>48.10±1.17</td>
<td>5.66±0.14</td>
<td>3.75±0.13</td>
</tr>
<tr>
<td>3</td>
<td>Cadmium chloride</td>
<td>27.51±2.18</td>
<td>3.41±0.24</td>
<td>2.39±0.19</td>
</tr>
<tr>
<td>4</td>
<td>Cadmium chloride + Green tea extract</td>
<td>35.23±1.70</td>
<td>4.04±0.13</td>
<td>2.01±0.17</td>
</tr>
</tbody>
</table>

Values are mean ± S.D for 6 rats in each group.

The significant decreases in Superoxide dismutase activities in liver was observed in group III rats when compared with group I rats, whereas group IV rats shows increases in the activities of SOD when compared with group III rats. There is no significant difference was observed in group II when compared with group I rat. Catalase activity was found to be significantly decreased in tissues of group III rats when compared with group I rats. Catalase activity was increased in group IV rats when compared with the corresponding group III rats. Group II rats showed no significant change in catalase activity when compared with group I rats. In
Glutathione peroxidase (GPx) significant decreases was observed in group II rats when compared with group I rats. Group IV rats showed significant activity in GPx when compared with group III rats. Group II rats showed no significant change when compared with group I rats.

**DISCUSSION**

Enhanced levels of TBARS in liver of Cadmium chloride (CdCl₂) treated rats indicated the increased levels of lipid peroxidation. Reports have shown that cadmium promotes the formation of ROS by Fenton transition equation, such as hydrogen peroxides and enhances the subsequent iron and copper-induced production of lipid peroxidations and the highly reactive hydroxyl radical [12-13]. Simultaneously administration of green tea extract decreased the formation of lipid peroxidation products, and it possesses antioxidant activity [14-15]. Thus, this agent might provide more medical benefit because the use of this agent could simultaneously alleviate oxidative damage [14]. The ability of green tea, consumed within a balanced controlled diet, to improve overall the antioxidants status and to protect against oxidative damage in humans [16]. Cadmium has a high affinity on GSH and causes the irreversible excretion of, upto two GSH tripeptides [17]. The metal-GSH conjugation process is desirable in that it results in the excretion of the toxic metal into the bile. However, it duplets the GSH from the cell and thus decreases the antioxidant potential [18]. SOD was observed in the liver of cadmium chloride intoxicated animals of our study are in agreement with the earlier reports of Iwalokun [19]. Catalase, which is present virtually in all mammalian cells, is responsible for the removal of Hydrogen peroxide. It plays an important role in the acquisition of tolerance to oxidative stress in adaptive response of cells. GSH is produced in the liver and maintained at a higher concentration in most tissues [20].

The decrease in the activities of antioxidant enzymes (SOD, CAT, GPx) in liver tissues of cadmium chloride treated rats may be due to the inhibition of these enzymes by H₂O and nitric oxide (NO). Both GSH and Cellular antioxidant enzymes plays an important role in CdCl₂ induced liver injury [21-22]. The metals inhibit the activites of SOD, CAT, and GPx in addition to the depletion of GSH content in tissues [23].

**CONCLUSION**

On the basis of the above results, it could be concluded that green tea (*Camellia sinesis*) possess significant antioxidant activity. It may be due to the presence of natural antioxidants and free radical scavenging activity.

**Acknowledgement**

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