ROLE OF MURRAYA KOENIGII (CURRY LEAF) AND BRASSICA JUNcea (MUSTARD) IN LIPID PEROXIDATION

BEENA A. KHAN, ANNIE ABRAHAM AND S. LEELAMMA*

Department of Biochemistry,
University of Kerala,
Kariavattom Campus,
Thiruvananthapuram - 695 581

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Abstract: The status of lipid peroxidation was investigated in rats fed M. Koenigii (curry leaf) and B. juncea (Mustard). Concentration of malondialdehyde showed a significant decrease, while hydroperoxides and conjugated dienes were significantly increased in liver and heart of both the experimental groups. SOD and catalase activity was found to be increased in liver and heart of both the spices administered groups. Glutathione levels in liver, heart and kidney were lowered in rats administered these spices. Glutathione reductase, glutathione peroxidase and glutathione S-transferase activity showed a sharp increase in the experimental groups compared to the controls.

Key words : murraya koenigii brassica juncea malondialdehyde superoxide dismutase glutathione s-transferase

INTRODUCTION

Curry leaf and mustard are extensively used in Ayurvedic medicines. Murray koenigii leaves, roots and bark are considered toxic stomachie and carminative. Leaves are used in dysentery and also for vomiting. Iyer and Mani (1) had shown the effect of curry leaf supplementation on lipid profile, glycated protein and aminoacids in noninsulin dependent diabetic patients. These beneficial effects are mediated in part, by inhibiting lipid peroxides and prostaglandin synthesis. However, the effect of spices on scavenging of reactive oxygen species has received very little attention. The present study was carried out to investigate the role of Murraya koenigii Spreng (Rutacea) and Brassica juncea (L) cosson (Cruciferae) on the metabolism of lipid peroxides in rats.

METHODS

Male albino rats of Sprague-Dawley strain weighing 80-85 g were divided into three groups of 6 each and were maintained on rat feed supplied by Hindustan Lever Limited.

Group I - Control
Group II - Diet mixed with 10% fresh curry leaves and powdered mustard seeds.

The rats were weighed every week up to a period of 60 days. After 18 hr fasting period the rats were stunned by a blow at the back of neck and killed by decapitation. The tissues (liver, heart, kidney) were removed to ice cold (0°C) containers for various estimations.

Estimation of enzymes activities: The following procedures were used for the estimation of various enzyme activities: Super oxide dismutase (SOD) (2) (EC 1.15.1.1) catalase (EC 1.11.1.6) (3). Glutathione reductase (GR,
EC 1.6.4.2) (4), Glutathione peroxidase (GPx, EC 1.11.1.9) (5), Glutathione S-transferase (GST, EC 2.5.1.18) (6), Glucose-5-phosphate dehydrogenase (G-6-PDH, EC 1.1.1.49) (7).

Malondialdehyde (MDA), hydroperoxides (HP) conjugated – dienes (CD) and free fatty acids (FFA) were monitored (8-11). Glutathione (12). Serum ceruloplasmin (13) and protein content (14) were estimated. Statistical analysis was done using the Student's 't' test.

RESULTS

The gain in weight was similar in control and experimental groups.

Malondialdehyde (MDA), concentration was decreased (P < 0.01) in liver and heart of both the experimental groups (Table IV). The concentration of hydroperoxides and conjugated – dienes increased (P < 0.01) in liver and heart of both the spices administered groups (Table IV). Free fatty acids showed a decrease (P < 0.01) in both the experimental groups in liver and heart (Table IV). The activity of superoxide dismutase and catalase was found to be increased (P < 0.01) in liver and heart of both the spices treated groups (Table III). Serum ceruloplasmin levels were not altered and glutathione levels in liver, heart and kidney showed a decrease (P < 0.01) in the animals fed M. koenigii leaves and seeds of B. juncea (Table I). Glutathione reductase, Glutathione peroxidase and glucose-6-phosphate dehydrogenase showed a sharp increase (P < 0.01) in their activities in the experimental groups, GST activity significantly increased (P < 0.01) in test groups (Table I, II).

TABLE I: Effect of curry leaf and mustard fed diet on serum ceruloplasmin levels, glutathione S-transferase and glutathione.
(The values are mean ± SEM of 6 rats)

<table>
<thead>
<tr>
<th>Ceruloplasmin (serum) (mg/dl/serum)</th>
<th>Control</th>
<th>17.2±0.39</th>
<th>Diet + curry leaves</th>
<th>17.6±0.26</th>
<th>Diet + mustard seeds</th>
<th>16.5±0.37</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td>45.2±</td>
<td>488.7±</td>
<td>494.4±</td>
<td>438.1±</td>
<td>405.7±</td>
<td>219.0±</td>
</tr>
<tr>
<td>Heart</td>
<td>11.8±</td>
<td>11.4±</td>
<td>3.9±</td>
<td>10.9±</td>
<td>10.7±</td>
<td>2.5±</td>
</tr>
<tr>
<td>Kidney</td>
<td>11.8±</td>
<td>11.4±</td>
<td>3.9±</td>
<td>10.9±</td>
<td>10.7±</td>
<td>2.5±</td>
</tr>
<tr>
<td>Glutathione (mM/100g)</td>
<td>11.8±</td>
<td>11.4±</td>
<td>3.9±</td>
<td>10.9±</td>
<td>10.7±</td>
<td>2.5±</td>
</tr>
<tr>
<td>Glutathione S-transferase (µmoles of thioester/min/mg/protein)</td>
<td>3.7±</td>
<td>4.4±</td>
<td>1.5±</td>
<td>4.9±</td>
<td>5.2±</td>
<td>1.9±</td>
</tr>
<tr>
<td></td>
<td>0.09</td>
<td>0.11</td>
<td>0.03</td>
<td>0.12</td>
<td>0.13</td>
<td>0.04</td>
</tr>
<tr>
<td>Glutathione reductase (mM NADPH oxidised/min/mg/protein)</td>
<td>16.9 ± 0.42</td>
<td>19.5 ± 0.48*</td>
<td>18.6 ± 0.46</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glutathione peroxidase (mg of reduced glutathione utilized/min/mg/protein)</td>
<td>5.9 ± 0.15</td>
<td>6.7 ± 0.16*</td>
<td>6.5 ± 0.16</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucose 6-phosphate dehydrogenase*</td>
<td>10.4 ± 0.28</td>
<td>12.9 ± 0.35*</td>
<td>12.7 ± 0.32*</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*P < 0.01.

TABLE II: Activity of glutathione reductase, glutathione peroxidase and glucose 6-phosphate dehydrogenase in the liver of rats fed with diet mixed with curry leaves and mustard seeds.
(The values are mean ± SEM of 6 rats)

| Glutathione reductase (mM NADPH oxidised/min/mg/protein) | Control | 16.9 ± 0.42 | Diet + curry leaves | 19.5 ± 0.48* | 18.6 ± 0.46 |
| Glutathione peroxidase (mg of reduced glutathione utilized/min/mg/protein) | 5.9 ± 0.15 | 6.7 ± 0.16* | 6.5 ± 0.16 |
| Glucose 6-phosphate dehydrogenase* | 10.4 ± 0.28 | 12.9 ± 0.35* | 12.7 ± 0.32* |

* P < 0.01 as compared to control
* (n moles of NADP+ red/min/mg protein)
TABLE III: Activity of superoxide dismutase (SOD) and catalase in the liver, heart and kidney of rats fed *Murraya koenigii* leaf and *Brassica juncea* seeds.

(Values are mean ± SEM of 6 rats)

<table>
<thead>
<tr>
<th>SOD</th>
<th>Liver</th>
<th>Heart</th>
<th>Kidney</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Group</strong></td>
<td><strong>I</strong></td>
<td><strong>II</strong></td>
<td><strong>III</strong></td>
</tr>
<tr>
<td><strong>I</strong></td>
<td>8.19 ± 0.204</td>
<td>13.71 ± 0.34</td>
<td>16.89 ± 0.42</td>
</tr>
<tr>
<td><strong>II</strong></td>
<td>17.76 ± 0.44</td>
<td>15.92 ± 0.39</td>
<td>22.47 ± 0.56</td>
</tr>
<tr>
<td><strong>III</strong></td>
<td>11.66 ± 0.29</td>
<td>16.28 ± 0.38</td>
<td>20.49 ± 0.51</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Catalase</th>
<th><strong>Group</strong></th>
<th>Liver</th>
<th>Heart</th>
<th>Kidney</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>I</strong></td>
<td>69.94 ± 1.7</td>
<td>9.8 ± 0.24</td>
<td>27.3 ± 0.68</td>
<td></td>
</tr>
<tr>
<td><strong>II</strong></td>
<td>87.2 ± 2.2</td>
<td>17.4 ± 0.43</td>
<td>40.7 ± 1.0</td>
<td></td>
</tr>
<tr>
<td><strong>III</strong></td>
<td>79.5 ± 1.9</td>
<td>15.8 ± 0.34</td>
<td>34.9 ± 0.87</td>
<td></td>
</tr>
</tbody>
</table>

**Catalase (x10^3 units **/mg protein)**

**Unit = Velocity constant/sec.**

Group I has been compared with group II and III.

DISCUSSION

The results show that feeding of curry leaf and mustard brings about alterations in the level of lipid peroxides in different tissues. The effect of spice principles on scavenging of reactive oxygen species has reported (15).

Maintenance of normal cell functions in presence of oxygen largely depends on the efficiency of the tissue protection against free radicals mediated oxidative stress. MDA, a breakdown product of unsaturated fatty acids seems to be decreased in the liver and heart of spice fed groups. The increased activity of enzymes in liver, kidney, heart and decreased levels of peroxides (TBARS) can result in decreased formation of toxic intermediates. The level of glutathione in tissues is altered in experimental groups. The tripeptide reduced glutathione is essential to maintain structural and functional integrity of the cells. The maintenance of GSH levels depends on the activities of various enzymes viz, glutathione reductase, glutathione s-transferase and glucose 6-phosphate.
dehydrogenase. Much of the $\text{H}_2\text{O}_2$ produced by the action of SOD is effectively eliminated from the system by another antiperoxidative enzyme glutathione peroxidase and catalase.

The increase in glucose 6-phosphate dehydrogenase leads to an increase in HMP shunt pathway and thereby increase the supply of NADPH. Such an increase in NADPH level resulted in the enhancement of glutathione reductase activity which in turn helps to keep a raised level of glutathione. Activity of GST, a multifunctional protein involved in the detoxification and possess peroxidase activity and participates in the reduction of fatty acid hydroperoxides to non-toxic alcohols (16) is significantly increased in our test groups agrees with these results. Similar increase in GST activity in nitrogen mustard (17), spices (18) and leafy vegetables (19) treated mice were reported. Serum ceruloplasmin levels were not altered in the spices fed groups. It has been reported that the ceruloplasmin can function as a pro or antioxidant depending on the ability of ceruloplasmin to influence the ratio of Fe (II) to Fe (III) by catalysing oxidation. The decreased levels of glutathione (reductase) should therefore result in decreased peroxidation.

The level of lipid peroxides in spices administered groups suggest that both curry leaf and mustard can maintain the levels of most of the biochemical parameters at the normal level.

REFERENCES