SHORT COMMUNICATION

ASCORBIC ACID, DEHYDROASCORBIC ACID AND GLUTATHIONE IN LIVER DISEASE

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Summary: Controlled studies were conducted to find out the plasma values of ascorbic acid, dehydroascorbic acid (DHA), urinary excretion of ascorbic acid and blood levels of glutathione in patients with viral hepatitis, alcoholic hepatitis, cirrhosis of liver and carcinoma of liver. Leucocyte ascorbic acid and DHA/AA index were also determined in order to assess the ascorbic acid status of these patients.

It was observed that the plasma and leucocytes contents of ascorbic acid were significantly subnormal with markedly decreased urinary excretion in these patients. Decreased level of glutathione and significantly higher level of DHA reflect an overall reducing status of the body is markedly deranged in these conditions. Further it was observed that the DHA/AA ratios were significantly altered in these groups of patients.

Key words: ascorbic acid, dehydroascorbic acid, glutathione, liver disease

INTRODUCTION

Several workers have reported that ascorbic acid, dehydroascorbic acid and glutathione values have considerable influence in regulating the overall reducing status of the body (2,7,8). Most of the investigators have studied either the leucocyte ascorbic acid level or plasma concentration in order to throw light on the ascorbic acid status of the patient suffering from various diseases (1).

Although Andrew's and Brooks (1) have recommended that measurement of leucocyte ascorbic acid reflects a true picture of ascorbic acid status of the patient, still a thorough
investigation regarding the plasma ascorbic acid, leucocyte ascorbic acid and urinary excretion of ascorbic acid is more meaningful (6,7,8). Such a comparison of data is necessary to distinguish between the dietary deficiency of ascorbic acid and the deficiency due to increased utilisation of ascorbic acid (7,8). Banerjee (2) has reported that the increase in serum DHA in diseased condition indicated increased catabolism of ascorbic acid. Further, he recommended that the measurement of DHA level along with leucocyte ascorbic acid and plasma ascorbic acid provides a clearer picture regarding the ascorbic acid status of the individual as well as the requirement of ascorbic acid under various diseased conditions. Further, Bhaduri et al. (5) have suggested that ascorbic acid and dehydroascorbic acid equilibrium is governed by glutathione to a great extent. Also, they have reported a significant decrease in the levels of ascorbic acid and GSH followed by a significant increase in the levels of DHA in patients of cholera, small pox, pyogenic meningitis, tuberculous meningitis, gonorrhoea and syphilis. Also, Banerjee (2) has reported that glutathione can protect ascorbic acid from oxidation to dehydroascorbic acid.

Work has been carried out on ascorbic acid deficiency in liver disease (4) but very limited literature is available on the reducing contents of the body with different liver disorders. The present study has been undertaken to evaluate the overall reducing status of the body with some liver disorders.

MATERIAL AND METHODS

The study is based on 64 subjects with liver disorders comprising 26 cases of viral hepatitis, 10 cases of alcoholic hepatitis, 17 cases of cirrhosis of liver and 11 cases of carcinoma of liver with 10 age matched individuals as control. Cases were selected from the gastroenterology clinic and the clinical diagnosis was confirmed by clinical evaluation, haematological investigations and histopathological examination.

Fasting blood was collected aseptically from antecubital vein in a heparinised tube. The plasma was assayed for dehydroascorbic acid and reduced ascorbic acid by 2:4 dinitrophenyl hydrazine method as described by Roe and Kuether (11). Assays were made within half an hour of the collection of the blood samples to prevent destruction of ascorbic acid. Leucocytes were separated by the method described by Gupta and Agarwal (9) and their ascorbic acid content was determined by 2:4 dinitrophenyl hydrazine method. Urine samples were collected and total ascorbic acid content was estimated by 2,6-dichlorophenol indophenol dye method as described by Varley (13). Glutathione was estimated as described by Beutler et al. (3). Ten normal individuals were selected and investigated for the above parameter to serve as controls.
RESULTS AND DISCUSSION

There was significant fall in the values of reduced ascorbic acid, leucocyte ascorbic acid, urinary excretion of ascorbic acid with a marked increase in the concentration of dehydroascorbic acid in all the groups of patients as compared to controls (Table I). The significant changes in the blood level of glutathione and DHA/AA ratio in these patients are shown in Table II. Marked decrease in the blood concentration of glutathione was found in these patients whereas the DHA/AA rations were significantly altered in all these patients.

TABLE I: Ascorbic acid status of the patients suffering from liver disorders.
(Results are expressed in terms of mean values±S. D.),

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Plasma AA (mg/100 ml)</th>
<th>Plasma DHA (mg/100 ml)</th>
<th>Leucocytes AA μg/10^6 cells</th>
<th>Urinary excretion of AA, mg/day</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.93 ±0.21</td>
<td>0.08 ±0.03</td>
<td>24.26 ±5.25</td>
<td>24.75 ±1.12</td>
</tr>
<tr>
<td>Viral hepatitis</td>
<td>0.48* ±0.18</td>
<td>0.51* ±0.14</td>
<td>13.09* ±2.25</td>
<td>12.22* ±1.19</td>
</tr>
<tr>
<td>Alcoholic hepatitis</td>
<td>0.47* ±0.15</td>
<td>0.52* ±0.16</td>
<td>14.18* ±3.99</td>
<td>14.83* ±1.40</td>
</tr>
<tr>
<td>Cirrhosis of liver</td>
<td>0.52* ±0.14</td>
<td>0.51* ±0.15</td>
<td>12.51* ±1.43</td>
<td>11.78* ±1.17</td>
</tr>
<tr>
<td>Carcinoma of liver</td>
<td>0.37* ±0.10</td>
<td>0.57* ±0.09</td>
<td>12.37* ±1.15</td>
<td>10.11* ±1.02</td>
</tr>
</tbody>
</table>

*P<0.001 : Figures in parantheses indicate number of cases.

It is interesting to note (Table I) that all the patients suffering from liver disorders included under the present study showed significantly decreased levels of leucocyte ascorbic acid. In addition all of them showed low concentration of plasma ascorbic acid and increased concentration of dehydroascorbic acid. The urinary excretion of ascorbic acid was also significantly decreased in these patients. These results together with the demonstration of increased plasma level of dehydroascorbic acid indicates that the patients are in poor state of ascorbic acid status which was maximum in cirrhosis of liver and carcinoma of liver.

This is in accordance with the observation of Krasner and Dymock (10) who further stated that ascorbic acid utilisation in patients with malignant disease was markedly increased and emphasised the ascorbic acid supplement to these patients. The observation that dehydroascorbic acid is markedly increased in all groups indicate that the catabolism of
ascorbic acid is markedly increased in these conditions. Now, it is evident from values in Table II that in all the groups of liver disorders, glutathione (GSH) values of blood were decreased and DHA/AA ratio, were altered. Since the glutathione contents of these patients are lower the protection of ascorbic acid from oxidation is decreased which results in an altered ratio of reduced ascorbic acid to oxidised ascorbic acid (2). This provides a plausible explanation for the significant increase observed in the plasma level of dehydro-ascorbic acid and altered ratio of DHA/AA in these patients.

**TABLE II**: Blood levels of glutathione and DHA/AA ratios in liver disorders.
(Results are expressed in terms of mean values ± S.D.)

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Glutathione (reduced) mg/100 ml blood</th>
<th>DHA/AA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>64.02 ± 4.83</td>
<td>0.1 ± 0.04</td>
</tr>
<tr>
<td>Viral hepatitis</td>
<td>43.06* ± 4.21</td>
<td>1.31* ± 0.85</td>
</tr>
<tr>
<td>Alcoholic hepatitis</td>
<td>41.25* ± 4.59</td>
<td>1.32* ± 0.81</td>
</tr>
<tr>
<td>Cirrhosis of liver</td>
<td>40.96* ± 6.59</td>
<td>1.1* ± 0.53</td>
</tr>
<tr>
<td>Carcinoma of liver</td>
<td>43.34* ± 4.29</td>
<td>1.49* ± 0.52</td>
</tr>
</tbody>
</table>

*P<0.001 : Figures in parantheses indicate number of cases.

From the above observations it can be safely concluded that the increased level of DHA is always associated with the decreased levels of glutathione, altered ratio of DHA/AA and with poor ascorbic acid status. It is difficult to conclude at this juncture that decreased levels of glutathione is due to diseased condition of the liver or due to poor availability of ascorbic acid but one point is obvious that these patients are in poor state of tissue status of ascorbic acid.

A clinical trial to explore the beneficial effect of ascorbic acid in these groups of patients may be undertaken to find out the place of mega doses of ascorbic acid as a therapeutic index.

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REFERENCES