Serum paraoxonase and arylesterase activities in metabolic syndrome in Zahedan, southeast Iran

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Abstract

Objective: Paraoxonase (PON) is associated with high-density lipoprotein and protects serum lipid from oxidation. The aim of this study was to determine serum PON, arylesterase (ARE) activities, and total antioxidant capacity (TAC) in metabolic syndrome (MES).

Methods: This case–control study was performed on 106 patients with MES and 231 healthy subjects. Serum PON and ARE activities were determined spectrophotometrically. TAC was determined using ferric reducing ability of plasma assay.

Results: The results showed that serum PON activity was significantly lower in patients with MES (69.62 ± 59.86 IU/l) than healthy subjects (91.64 ± 77.45 IU/l) (P < 0.05). The serum ARE activity in MES and normal subjects were 45.23 ± 23.24 and 65.69 ± 31.10 kU/l respectively. The ARE activity was significantly lower in patients with MES than normal subjects (P < 0.0001). No significant differences were observed between MES and normal subjects regarding TAC.

Conclusion: The lower PON and ARE activities in MES may be considered an independent risk factor for cardiovascular disease, which remains to be cleared.

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Introduction

Metabolic syndrome (MES), a collection of cardiovascular risk factors including central obesity, hypertension, hyperglycemia, glucose intolerance, and dyslipidemia, is associated with an increased risk of cardiovascular disease and diabetes (1). Human serum paraoxonase 1 (PON1) is ~43–45 kDa glycoprotein, synthesized mainly by the liver, which circulates in serum in association with high-density lipoprotein (HDL) and protects low-density lipoprotein (LDL) from oxidation by the hydrolysis of biologically active lipoperoxides (2). The activity of this enzyme is measured using paraoxon or is estimated from the activity of arylesterase (ARE) using phenyl acetate. It has been reported that ARE activity is not affected by the polymorphisms of PON1 (3, 4).

Serum PON1 activity was found to be reduced in a number of pathological conditions including coronary artery disease (5), hypercholesterolemia (6), type 2 diabetes (6, 7), polycystic ovary syndrome (8), and renal failure (9). PON1 is recognized as an antioxidant enzyme because it hydrolyzes lipid peroxides in oxidized lipoproteins.

To the best of our knowledge, information regarding PON and ARE activities in MES is limited. The aim of this study was to find out the levels of PON and ARE activities in MES.

Materials and methods

This case–control study was performed on 106 individual with MES and 231 normal subjects. The demographic and biochemical characteristic of the groups are shown in Table 1. The study was approved by the local ethical committee of Zahedan University of Medical Sciences and written informed consent was obtained from all subjects. The MES was determined as the presence of three or more of five components according to the national cholesterol education program (NCEP) ATP III (Table 2) (1).

Fasting blood glucose (FBG) and lipid profile (TG, total cholesterol, LDL-C, and HDL-C concentrations) were measured by automated chemistry analyzer using commercial available kits (Table 1).
PON activity assays were performed in the absence (basal activity) and presence of 1 M NaCl (salt-stimulated activity) using paraoxon (diethyl-p-nitrophenyl phosphate) as a substrate as described previously (10). Phenylacetate was used as a substrate to determine the ARE activity. The rate of phenol produced was continuously monitored at 270 nm at 37 °C. ARE activity was determined using molar extinction coefficient of phenol (1310/M per cm) and expressed as kU/l serum (10).

Serum total antioxidant capacity (TAC) was determined by measuring their ability to reduce Fe3+ to Fe2+, which is known as ferric reducing ability of plasma (FRAP) assay as described previously (11).

Statistical analysis was performed by commercial software (SPSS for Windows, V17) using independent sample t-test and the Pearson correlation coefficient test. A P value <0.05 was considered statistically significant.

### Table 1 Biochemical parameters in metabolic syndrome (MES) and normal subjects.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>MES</th>
<th>Normal subjects</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>FBG (mg/dl)</td>
<td>118.50 ± 58.71</td>
<td>85.42 ± 12.33</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TG (mg/dl)</td>
<td>222.89 ± 190.66</td>
<td>113.33 ± 48.31</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>212.71 ± 48.80</td>
<td>173.71 ± 40.25</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>HDL-C (mg/dl)</td>
<td>41.58 ± 8.00</td>
<td>45.14 ± 6.89</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>125.9 ± 45.17</td>
<td>102.73 ± 34.23</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>160.71 ± 9.70</td>
<td>163.46 ± 10.17</td>
<td>0.02</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>75.39 ± 14.02</td>
<td>63.72 ± 13.03</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>99.32 ± 12.41</td>
<td>82.39 ± 15.77</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>125.89 ± 85.89</td>
<td>102.73 ± 34.23</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>58.71 ± 8.00</td>
<td>45.14 ± 6.89</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

### Results

The study consisted of 106 MES (34 males and 72 females; age 43.54 ± 14.07) and 231 normal subjects (97 males and 134 females; age 35.64 ± 13.27). The levels of PON1 activity in MES and healthy subjects were 69.62 ± 59.86 and 91.64 ± 77.45 IU/l respectively. The activity of PON in MES was significantly lower than normal subjects (P<0.01). In addition, salt-stimulated PON activity was significantly lower in MES (136.23 ± 111.80 IU/l) than normal subjects (192.24 ± 162.68 IU/l) (P<0.02).

The serum ARE activity in MES and normal subjects were 45.23 ± 23.24 and 65.69 ± 31.10 kU/l respectively (Table 3).

In males, there were no significant differences in the activities of PON1 between MES (64.57 ± 58.24 IU/l) and normal subjects (90.01 ± 84.17 IU/l) (P=0.106). While a significant difference was found regarding ARE activity in MES (64.68 ± 35.08 kU/l) and normal subjects (40.97 ± 16.23 kU/l) (P<0.001).

In females, the activity of PON was significantly lower in the MES (72.01 ± 60.87 IU/l) than in the controls (92.82 ± 72.49 IU/l) (P=0.039). We also found that ARE activity was significantly lower (P<0.001) in female cases (47.25 ± 25.75 kU/l) than in normal subjects (62.80 ± 27.6 kU/l).

No significant correlation was observed between age and PON or ARE activities (P>0.05). A significant difference was observed among MES and normal subjects regarding the ARE activity (P<0.0001). No significant difference was observed among MES and normal subjects for TAC. In MES and normal subjects, PON activity was positively correlated with ARE activity (r=0.368, P<0.0001; r=0.594, P<0.0001). While a positive correlation was observed between PON and HDL-C in normal subjects (r=0.168, P=0.01), the correlation between MES and HDL-C was not significant (r=0.122, P=0.21). Furthermore, there were no correlations between MES and normal subjects regarding PON and cholesterol, LDL-C, triglyceride, TAC, and body mass index (BMI) (P>0.05).

### Discussion

In this study, we found that PON and ARE activities were significantly lower in MES when compared with normal subjects. The formation of free radicals is a normal outcome of a variety of essential biochemical reactions and can occur at elevated rates under pathophysiological circumstances (12, 13). The physiological role of antioxidants is to prevent damage to cellular components and DNA. The diminished PON and ARE activities in MES suggest an increased risk of oxidative stress which may be associated with the development of the metabolic syndrome.

### Table 2 Criteria for diagnosis of the metabolic syndrome according to the national cholesterol education program (NCEP) ATP III.

<table>
<thead>
<tr>
<th>Criterion</th>
<th>NCEP ATP III</th>
</tr>
</thead>
<tbody>
<tr>
<td>Waist (cm)</td>
<td>≥ 102</td>
</tr>
<tr>
<td>HDL (mg/dl)</td>
<td>≥ 88</td>
</tr>
<tr>
<td>Male</td>
<td>&lt; 40</td>
</tr>
<tr>
<td>Female</td>
<td>&lt; 50</td>
</tr>
<tr>
<td>Triglyceride (mg/dl)</td>
<td>≥ 150</td>
</tr>
<tr>
<td>Fasting glucose (mg/dl)</td>
<td>≥ 100</td>
</tr>
<tr>
<td>Blood pressure (mmHg)</td>
<td>≥ 130/85</td>
</tr>
</tbody>
</table>

*Three of five required.

### Table 3 The levels of paraoxonase (PON), salt-stimulated PON, arylesterase activities, and total antioxidant capacity (TAC) in metabolic syndrome (MES) and healthy subjects.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>MES</th>
<th>Normal subjects</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PON1 activity (IU/l)</td>
<td>69.62 ± 59.86</td>
<td>91.64 ± 77.45</td>
<td>0.01</td>
</tr>
<tr>
<td>Salt-stimulated PON (IU/l)</td>
<td>136.23 ± 111.80</td>
<td>192.24 ± 162.68</td>
<td>0.02</td>
</tr>
<tr>
<td>ARE (kU/l)</td>
<td>45.23 ± 23.24</td>
<td>65.69 ± 31.10</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TAC (μmol/l)</td>
<td>972.46 ± 374.13</td>
<td>966.97 ± 380.60</td>
<td>0.912</td>
</tr>
</tbody>
</table>
cellular components arising as a consequence of chemical reactions involving free radicals. PON1 is a specific antioxidative enzyme with both PON and ARE activities. There is little information regarding the levels of PON and ARE in MES. Senti et al. (14) had found that the serum PON1 activity was significantly lower in MES than in normal subjects. Our results are in agreement with this finding. Tabur et al. (15) had found that the levels of PON and ARE activities were not significantly different among non-diabetic MES, non-MES obese patients, and healthy subjects, while total antioxidant status was low in both the MES and obese groups compared to controls.

A variety of PON1 gene polymorphisms have been recognized (16, 17). It has been well documented that the two common coding region polymorphisms of the gene PON1 (L55M and Q192R) lead to changes of both the level and activity of the enzyme (18, 19). Moreover, it has been found that promoter polymorphism of PON1, especially −1077T/C, affects the PON1 expression and serum concentration (20). On the other hand, acquired factors such as diseases, diet, and lifestyle can also affect the PON1 activity. It has been proposed that consumption of red wine or flavonoid-containing drinks (21) as well as moderate alcohol intake (22) increases serum PON1 activity. The exact mechanisms affecting low PON1 and ARE activities in MES are yet to be clear.

It is well known that in the general population, MES is associated with increased cardiovascular morbidity and mortality (23, 24) and high prevalence of type 2 diabetes mellitus (25). Human PON1, an HDL-associated enzyme, is capable of preventing LDL oxidation. According to this and previous results (14), reduced PON and ARE activities in MES might be an independent risk factor for cardiovascular disease in these patients. However, more studies in diverse populations are needed for clear confirmation.

Declaration of interest
The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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