Serum Dehydroepiandrosterone Sulphate Levels in Patients with Non-Alcoholic Fatty Liver Disease

Masafumi Koga¹, Hiroshi Saito¹, Mikio Mukai¹, Toshiji Saibara² and Soji Kasayama³

Abstract

Background  Dehydroepiandrosterone (DHEA) is an adrenal hormone reported to prevent body weight gain, diabetes mellitus and atherosclerosis. We hypothesized that DHEA is involved in the pathophysiology of non-alcoholic fatty liver disease (NAFLD) often associated with obesity and insulin resistance. In this study, we aimed to examine the clinical significance of serum DHEA sulfate (DHEAS) in patients with NAFLD.

Methods  We determined serum DHEAS, serum alanine aminotransferase (ALT), serum lipids, plasma fasting glucose and insulin levels in 158 Japanese men who had neither viral hepatic diseases nor alcohol intake exceeding 20 g/day. NAFLD was diagnosed by the presence of fatty change of the liver by echotomographic examination.

Results  Among the study subjects, 69 were diagnosed as having NAFLD. Their serum DHEAS levels were significantly higher than in 89 subjects without NAFLD. Serum DHEAS levels in 19 NAFLD patients with elevated ALT levels (>40 U/L) were significantly higher than in the other 50 NAFLD patients with normal ALT levels (≤40 U/L). Multivariate regression analysis demonstrated that serum ALT was positively correlated with serum DHEAS, serum triglyceride and body mass index.

Conclusion  Serum DHEAS levels are increased in patients with NAFLD with elevated ALT levels. Increased serum DHEAS may be a component of the pathophysiology of NAFLD.

Key words: dehydroepiandrosterone, non-alcoholic fatty liver disease, fatty liver, alanine aminotransferase, non-alcoholic steatohepatitis

NASH. The aim of this study was to analyze the clinical significance of DHEAS levels in patients with NAFLD.

Table 1. Clinical Characteristics of the Subjects with or without NAFLD

<table>
<thead>
<tr>
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<th>NAFLD</th>
<th>–</th>
<th>+</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>69</td>
<td>89</td>
<td></td>
<td></td>
</tr>
<tr>
<td>age (years)</td>
<td>50.0±7.0</td>
<td>51.5±6.9</td>
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<tr>
<td>BMI (kg/m²)</td>
<td>26.6±3.4</td>
<td>22.8±2.5</td>
<td>&lt;0.001</td>
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<tr>
<td>serum AST (U/L)</td>
<td>27±13</td>
<td>19±4</td>
<td>&lt;0.001</td>
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<tr>
<td>serum ALT (U/L)</td>
<td>27±13</td>
<td>20±6</td>
<td>&lt;0.001</td>
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<tr>
<td>serum triglyceride (mg/dL)</td>
<td>197±99</td>
<td>116±53</td>
<td>&lt;0.001</td>
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<tr>
<td>serum HDL cholesterol (mg/dL)</td>
<td>48±11</td>
<td>55±14</td>
<td>&lt;0.001</td>
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<tr>
<td>serum LDL cholesterol (mg/dL)</td>
<td>126±25</td>
<td>125±26</td>
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<td></td>
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<tr>
<td>fasting plasma glucose (mg/dL)</td>
<td>109±23</td>
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<td>HOMA-%S (%)</td>
<td>109±52</td>
<td>187±80</td>
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<tr>
<td>DHEAS (ng/mL)</td>
<td>1,814±842</td>
<td>1,646±661</td>
<td>0.004</td>
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</table>

Materials and Methods

Subjects

We initially recruited a total of 278 Japanese men who visited the Health Care Center at Kinki Central Hospital for health examinations. Of these subjects we excluded subjects with viral hepatic diseases (positive hepatitis B surface antigen test, positive hepatitis C antibody test and/or past histories of hepatitis B or hepatitis C), diabetes mellitus, renal disease and malignant disease. Subjects who ingested daily alcohol exceeding 20 g/day and subjects who had serum ALT>40 U/L without fatty change of the liver were also excluded. A total of 158 men satisfied the above admission criteria. Their average age was 50.8±7.0 years and body mass index (BMI) was 24.4±3.5 kg/m².

Laboratory methods

Fasting plasma glucose, serum insulin, serum alanine aminotransferase, serum total cholesterol, high density lipoprotein (HDL) cholesterol and serum triglycerides were determined by means of standard laboratory assays. Serum DHEAS was measured by radioimmunoassay (DPC DHEAS kit, Mitsubishi Kagaku Iatron, Tokyo, Japan). Inter- and intra-assay coefficients of variation in this assay were 12.0% and 4.1% at the concentrations of 600 and 650 ng/mL (14). The homeostasis model assessment insulin sensitivity index (HOMA-%S) was calculated from fasting plasma glucose and serum insulin concentrations by using the correct HOMA evaluation and a computer program (15).

Fatty change of the liver was evaluated by echotomographic examination by assessing hepato-renal contrast, impaired visualization of the hepatic vein borders or the diaphragm (16). When the fatty change was observed in the study patients, they were diagnosed to have NAFLD. Furthermore, we judged the degree of fatty liver as moderate or more when the findings of impaired visualization of the hepatic vein borders or the diaphragm were observed. Otherwise we judged as mild degree of fatty liver.

Statistical analyses

Data are shown as means ± SD. Comparisons of serum DHEAS levels between groups were analyzed with a Student’s t-test. Univariate as well as stepwise multivariate regression analyses were performed to assess the effects of variables on serum ALT levels. For the stepwise multivariate regression analyses, the F value for the inclusion of the variables was set at 4.0. In the univariate as well as multivariate regression analyses, serum ALT and serum DHEAS were logarithmically transformed, to correct for skewed distribution. These analyses used a StatView computer program (Version 5.0). The statistical differences were considered to be significant at p<0.05.

Results

Of the 158 study subjects, 69 were diagnosed as having NAFLD, whereas the other 89 were not. In the patients with NAFLD BMI, AST, ALT, triglycerides and low density lipoprotein (LDL) cholesterol were significantly higher than in the subjects without it (Table 1). HDL cholesterol and HOMA-%S were significantly lower in the patients with NAFLD than in the subjects without it. Serum DHEAS levels were on average 1,810±840 ng/mL in the patients with NAFLD, which was significantly higher (p=0.0039) than those in the subjects without it (1,460±660 ng/mL) (Table 1, Fig. 1A). There was a significant difference between serum DHEAS levels in 29 subjects with a mild degree of fatty liver and in 43 subjects with moderate or more degree of fatty liver (1,890±904 ng/mL vs. 1,769±810 ng/mL; p=0.5660). Serum DHEAS levels were not associated with BMI (R=0.160, p=0.0524) or fasting plasma glucose (R=0.022, p=0.7819), while they showed a weak but significant association with body weight (R=0.155, p=0.0443) and HOMA-%S (R=0.180, p=0.0216). Among 69 patients with NAFLD, 19 had elevated ALT levels (>40 U/L). Their serum DHEAS levels were 2,380±950 ng/mL, which were significantly higher (p<0.0001) than those in the other 139 subjects with normal ALT levels (≤40 U/L) (1,510±670 ng/mL) (Fig. 1B). They were also significantly higher (p=0.0003) than those in 50 NAFLD patients with normal ALT levels (1,600±690 ng/mL). Serum ALT levels were associated positively with serum triglycerides (R=0.399, p<0.0001), BMI (R=0.418, p<0.0001), serum DHEAS (R=0.322, p<0.0001) and fasting plasma glucose (R=0.183, p=0.0204), and inversely with HOMA-%S (R=-0.419, p<0.0001), age (R=-0.224, p=0.0043) and serum HDL cholesterol (R=-0.186, p=0.0183) in the 160 study subjects (Ta-
Table 2 and Fig. 2). Stepwise multivariate regression analysis showed that serum DHEAS in addition to serum triglycerides and BMI were independently associated with serum ALT (Table 2).

Discussion

In the present investigation, we examined serum DHEAS levels in 158 men without viral hepatic diseases, diabetes mellitus, renal disease, malignant disease, or excess alcohol drinking habits. Since serum DHEAS levels significantly differ between men and women (17), this study was performed in men. The results clearly showed independent association of serum DHEAS levels with serum ALT levels. Serum DHEAS levels were significantly higher in patients with NAFLD than in those without it. In addition, serum DHEAS levels were higher in who had elevated serum ALT levels. Thus, it is suggested that serum DHEAS levels increase in patients with NAFLD with high levels of serum ALT.

Serum DHEAS levels depend on adrenal DHEA production and its hepatic metabolism mediated by DHEA sulfotransferase catalyzing sulfonation of DHEA to form DHEAS. Although the activity and concentration of DHEA sulfotransferase has been shown to be reduced in hepatic tissues derived from primary biliary cirrhosis, primary sclerosing cholangitis, chronic active hepatitis and alcoholic cirrhosis compared with normal liver (18), there is no report on the content and activity of DHEA sulfotransferase in that derived from NAFLD. Thus, it is not known whether or not the increased serum DHEAS levels in NAFLD result from the increased sulfonation of DHEA.

Yoneda et al (19) studied the effects of DHEA treatment on hepatic injury by concanavalin A-induced T lymphocytes in mice. They showed that DHEA reduced hepatic injury by inhibiting several inflammatory mediators such as tumor necrosis factor α and macrophage mitogen inhibitory factor, and prevented the increase of serum ALT levels. Thus DHEA might have a protective effect against hepatotoxicity in this mouse model. Although the roles of DHEA in NAFLD have not yet been proven, the increased serum DHEAS levels in patients with NAFLD may reflect a compensatory increase of DHEA production to protect against hepatic damage. Recently, it has been reported that serum DHEAS levels were rather decreased in patients with histologically diagnosed NASH (12, 13). They have also shown that serum DHEAS levels were lower in NASH patients with NAFLD.
with incremental fibrosis stage (12, 13). In the present investigations, we failed to perform histological diagnosis of the liver of patients with NAFLD and thus it was not determined how many of these patients represented NASH. We, however, propose that most of them did not suffer from NASH since the frequency of NASH patients among NAFLD patients is suggested to be around 10 % (20). Increased DHEAS in patients with NAFLD may reflect increased adrenal secretion of DHEA for the prevention of the development and progression of hepatic damage (hepatoadrenal axis), while reduced DHEAS levels in patients with advanced stage of NASH may result mainly from the reduced sulphonation of DHEA. Further clinical and experimental studies are necessary to prove these hypotheses.

There is an increasing number of reports demonstrating that insulin resistance and the metabolic syndrome are involved in the development and progression of NAFLD (5, 9). These are also supported by our results in that the insulin sensitivity index HOMA-%S was inversely associated with serum ALT. DHEA is known to have potential to improve insulin sensitivity in vivo (21-24). It also has properties to increase insulin sensitivity in hepatocytes (25). In addition, it has been shown that DHEA can inhibit 11β-hydroxysteroid dehydrogenase 1 expression in liver and adipose tissues (26). It is proposed that the overexpression of 11β-hydroxysteroid dehydrogenase 1 in adipose tissues increases the local cortisol production and thereby leads to the metabolic syndrome-like status (27, 28). In the present study, serum DHEAS was a factor associated with serum ALT, independent of BMI, HOMA-%S and serum triglycerides. Therefore, DHEAS may be a component of NAFLD, in addition to obesity and its related metabolic disorders.

**The authors state that they have no Conflict of Interest (COI).**

<table>
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<th>Variable</th>
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<th>p value</th>
<th>Partial regression coefficient</th>
<th>F</th>
<th>p value</th>
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Explanatory variables in multivariate regression analysis included are age, BMI, serum triglyceride, serum high density lipoprotein (HDL) cholesterol, serum low density lipoprotein (LDL) cholesterol, fasting plasma glucose, HOMA-%S and serum DHEAS. R²=0.307, F=22.4, and p<0.0001.

## References