INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) is a clinicopathological syndrome characterized by hepatic steatosis with or without active inflammation (1) in patients with a negligible alcohol intake. There is growing concern about NAFLD, not only because this is a common liver disorder with a worldwide distribution, but also because it is recognized as one of the leading causes of chronic liver disease (2). In addition, a recent study has revealed that patients with nonalcoholic steatohepatitis (NASH) may progress to liver fibrosis, and approximately 8-17% progress to cirrhosis (2, 3). Although NAFLD may occur in non-obese patients (1), most cases of NAFLD are associated with obesity, type 2 diabetes mellitus (4), and hyperlipidemia (5). Weight reduction alone can improve liver function in obese patients with fatty liver (6). Moreover, insulin resistance underlies most cases of NAFLD, using the homeostasis model assessment-insulin resistance (HOMA-IR) method (7, 8), with a resultant increase in circulating insulin levels (9).

Adiponectin is a 30-kDa protein (10). In normal humans, its expression is restricted to adipose tissue (11). Plasma adiponectin levels are negatively correlated with the body mass index (BMI), fasting plasma glucose, fasting insulin, insulin resistance, and triglycerides (12). It is an anti-inflammatory adipocytokine that modulates insulin effects (13). The administration of adiponectin to mice decreased the plasma glucose (10), free fatty acid (FFA) and triglyceride levels (14), and hepatic glucose production (13). Plasma adiponectin levels are directly correlated with insulin sensitivity and, consequently, with decreases in obese and type 2 diabetic patients (11, 15). Since adiponectin appears to induce insulin sensitivity, we hypothesized that hypoadiponectinemia is associated with NAFLD. Therefore, we investigated the relationship between NAFLD and plasma adiponectin levels and insulin resistance.

MATERIALS AND METHODS

The study subjects were recruited from participants in routine health examinations at the Department of Family Medicine, Korea University Hospital, Seoul, Korea, in February 2004. The study was approved by the ethics committee of Anam Hospital, and was conducted in conformity with the
Table 1. Anthropometric and metabolic variables of NAFLD and control groups in men and women, respectively

| Variables                  | Control       | NAFLD         | p value | Control       | NAFLD         | p value |
|----------------------------|---------------|---------------|---------|---------------|---------------|---------|
| Number (%)                 | 15 (58)       | 11 (42)       |         | 38 (58)       | 27 (42)       |         |
| Age (yr)                   | 45.3±11.7     | 51.8±6.5      | 0.33    | 49.3±8.4      | 54.2±8.2      | 0.19    |
| BMI (kg/m²)                | 23.7±1.8      | 24.5±2.1      | <0.01   | 26.0±2.1      | 27.1±3.5      | <0.01   |
| Waist circumference (cm)   | 85.3±5.7      | 81.6±6.9      | 0.02    | 83.6±12.6     | 88.6±8.7      | 0.01    |
| Body fat mass (%)          | 19.0±3.2      | 31.5±4.3      | <0.01   | 23.5±4.5      | 34.4±5.8      | 0.03    |
| Systolic BP (mmHg)         | 122.1±21.5    | 118.5±10.1    | 0.96    | 122.5±13.6    | 124.8±14.8    | 0.05    |
| Diastolic BP (mmHg)        | 72.5±14.1     | 74.5±9.9      | 0.15    | 79.6±9.25     | 79.3±9.1      | 0.05    |
| Total cholesterol (mg/dL)  | 199.4±43.3    | 207.8±42.3    | 0.62    | 198.3±35.3    | 204.6±31.4    | 0.45    |
| HDL-cholesterol (mg/dL)    | 46.6±10.4     | 41.1±6.7      | 0.12    | 57.1±17.1     | 49.4±12.9     | 0.04    |
| LDL-cholesterol (mg/dL)    | 124.9±32.8    | 129.3±39.6    | 0.76    | 117.0±28.7    | 127.3±24.7    | 0.14    |
| Triglycerides (mg/dL)      | 137.8±78.7    | 204.7±46.4    | 0.01    | 133.2±70.7    | 159.8±74.5    | 0.15    |
| AST (U/L)                  | 19.4±4.8      | 22.5±4.3      | 0.10    | 23.6±18.3     | 25.9±9.1      | 0.55    |
| ALT (U/L)                  | 21.4±7.3      | 26.7±10.1     | 0.05    | 23.2±30.5     | 32.0±19.0     | 0.15    |
| Fasting blood glucose (mg/dL) | 94.3±11.4   | 93.2±11.8 | 0.84  | 89.9±9.1 | 96.1±16.1 | 0.05  |
| Fasting insulin (µU/mL)   | 5.92 (5.20-8.17) | 8.19 (7.00-9.43) | 0.04 | 6.61 (5.84-8.83) | 9.01 (7.71-13.90) | <0.01 |
| HOMA-IR                   | 1.40 (1.17-1.80) | 1.99 (1.56-2.40) | 0.04 | 1.56 (1.24-2.01) | 2.31 (1.69-3.23) | <0.01 |
| Adiponectin (µg/mL)       | 5.58 (3.92-8.70) | 2.50 (2.02-3.84) | <0.01 | 8.38 (5.15-12.11) | 6.17 (3.69-9.52) | <0.01 |

Data are expressed as mean±SD for Gaussian variables and median and lower and upper quartiles for non-Gaussian variables.

NAFLD, non-alcoholic fatty liver disease; BMI, body mass index; BP, blood pressure; HDL-cholesterol, high-density lipoprotein cholesterol; LDL-cholesterol, low density lipoprotein cholesterol; AST, aspartate aminotransferase; ALT, alanine aminotransferase; HOMA-IR, homeostasis model of assessment insulin resistance.
presence of NAFLD as the dependent variable, and adiponec-
tin, BMI, waist circumference, HOMA-IR, age, and sex as
independent variables. The level of statistical significance was
set at \( p < 0.05 \). All statistical analyses were carried out using the
SAS computer analysis program (version 8.2; SAS Institute).

**RESULTS**

Anthropometric and metabolic characteristics of NAFLD
and control groups in men and women, respectively, in Table
1. The study subjects were included 26 men and 65 women.
The mean age was 51.3 ± 8.8 yr and the mean BMI was 25.3
± 2.9 kg/m². There was no significant difference in plasma
adiponectin levels as well as other clinical parameters between
control group and non-participants of this study. NAFLD
subjects had significantly higher BMI, waist circumference,
fat mass, fasting insulin, and HOMA-IR, and had significantly
lower plasma adiponectin than control groups. Log levels of
plasma adiponectin were significantly lower in men than in
women (0.50 ± 0.35 and 1.34 ± 0.54, respectively, \( p < 0.001 \)).

Fig. 1 indicates plasma adiponectin levels and HOMA-IR
in the NAFLD and control groups in men and women, respecti-
vely. Plasma adiponectin levels in NAFLD group were sig-
nificantly lower than those in control group in both men and
women. HOMA-IR in NAFLD group were significantly
higher than those in control group in both men and women.

Fig. 2 graphically depicts the correlations of plasma adipon-
ectin levels with HOMA-IR and waist circumference accord-
ing to sex. Plasma adiponectin levels were significantly inverse
correlated (\( r = -0.45, p < 0.01 \)) with HOMA-IR in women. In
the other hands, the associations of plasma adiponectin levels
with waist circumference was significantly inverse correlated
in both men and women (\( r = -0.35, p = 0.03, r = -0.41, p < 0.01, \)
respectively)

The univariate correlations and partial correlation analyses
after adjusting for age, sex, and adiposity (BMI, waist circum-
ference, and fat mass) between plasma adiponectin levels and
anthropometric and metabolic parameters are shown in Table
2. Adiponectin levels correlated with waist circumference
DISCUSSION

We found that the NAFLD group had a higher insulin resistance than the control group, which is in agreement with previous studies (17, 18). NAFLD is associated with insulin resistance and hyperinsulinemia in even lean subjects with normal glucose tolerance (18). It was suggested that insulin resistance is the pathognomonic condition responsible for NAFLD. Indeed, NAFLD is considered the hepatic manifestation of metabolic syndrome.

Insulin resistance is an essential requirement of NASH, independent of the degree of obesity (12). The insulin-sensitizing drugs troglitazone (19) and metformin (20) reduce aminotransferase levels. In the insulin-resistant state, accelerated lipolysis of adipose tissue results in an increased supply of hepatic free fatty acids (FFAs) and increased lipid oxidation, this is accompanied by fat accumulation in hepatocytes. There is a good correlation between the liver fat content and liver insulin resistance in normal subjects and in type 2 diabetic patients (20).

In this study, insulin resistance was measured using the homeostasis model assessment method, although euglycemic-hyperinsulinemic clamp is the gold standard for defining insulin resistance (7, 8). However, HOMA-IR is easy to perform, and that method is highly correlated with the euglycemic-hyperglycemic clamp (r=0.83; p<0.01) (21). The HOMA method for measuring insulin resistance has been applied extensively in epidemiological investigations (7, 8, 21).

Our study confirms that hypoadiponectinemia occurs in subjects with NAFLD, after controlling for age, sex, and adiposity. Animal models have indicated that adiponectin confers protective effects against alcoholic and nonalcoholic fatty liver disease (22, 23). Recent study reported that hypoadiponectinemia is a feature of NASH independent of insulin resistance and reduced adiponectin level is associated with more extensive necroinflammation and may contribute to the development of necroinflammatory forms of NAFLD (24).

These data might also support the hypothesis that adiponectin has hepatoprotective effects in humans with NAFLD. The most likely reason for low adiponectin levels in NAFLD may be insulin resistance.

Our study found that HOMA-IR was significantly negatively correlated to adiponectin levels which were in accord with a previous report (25). Many investigators (8, 26, 27) suggest that adiponectin regulates hepatocyte metabolism.
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directly. Long-term administration of adiponectin to diabetic mice improved the indices of insulin sensitivity, and decreased liver, muscle, and plasma triglycerides, and FFAs (22). Injection of recombinant adiponectin in mice increases fatty acid oxidation in muscle, reduces triglyceride content in muscle, and improves muscle sensitivity to insulin (14).

Raised plasma tumor necrosis factor (TNF)-α is thought to be another reason for the low adiponectin levels in NAFLD (28). Overproduction of the proinflammatory cytokine TNF-α by adipose tissue is involved in insulin resistance in obesity and TNF-α is a major cytokine contributing to liver damage in NAFLD (29).

In our study, plasma adiponectin levels were elevated more in women than in men. Similar results have been reported in several studies (29, 30), while others failed to observe a sex difference (31). The higher adiponectin expression in women, as compared to men, might be due to the fact that women tend to have less visceral fat tissue than subcutaneous fat tissue. Plasma adiponectin levels were determined predominantly by visceral fat, not by subcutaneous fat (32). Therefore, sexual dimorphism of the body fat distribution might contribute to the difference in plasma adiponectin levels in women and men.

Among the anthropometric index, waist circumference was stronger correlation with plasma adiponectin level, in our study. According to definition for metabolic syndrome suggested by the National Cholesterol Education Program Adult Treatment Panel III (33), abdominal obesity is one of the diagnostic criteria of the metabolic syndrome. Increased adiposity in the abdominal area may result in the development of insulin resistance, so abdominal obesity estimated by waist circumference is inverse correlation with plasma adiponectin levels.

This study has some limitations. One limitation of this study is that the diagnosis of NAFLD was based on ultrasound examination, but was not confirmed pathologically. It is difficult to perform extensively in subjects with NAFLD, but was not confirmed pathologically.

Another limitation is that the diagnosis of NAFLD was based on ultrasonography, but was not confirmed pathologically. It is difficult to perform extensively in subjects with NAFLD, but was not confirmed pathologically. Therefore, our study had a cross-sectional design, and there was potential bias in participation, so it cannot elucidate mechanisms or determine the direction of causality.

In conclusion, we demonstrated that hypoadiponectinemia and insulin resistance are associated with NAFLD independent of total heaviness and abdominal fat distribution. Adiponectin may be applicable in human disease as a novel agent for treating insulin resistance including NAFLD in the future. Further research is needed to identify the key determinants of circulating adiponectin and the development of NAFLD.

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