ORIGINAL

A/G heterozygote of the A-3826G polymorphism in the UCP-1 gene has higher BMI than A/A and G/G homozygote in young Japanese males

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Abstract : UCP-1 is suggested to have important roles for thermogenesis and energy expenditure. To elucidate whether the A-3826G polymorphism that is located in the 5' flanking region of the UCP-1 gene has roles in healthy young people, the polymorphism was genotyped among 251 young Japanese men whose mean age is 22.7 years old. We analyzed relationship between the A-3826G polymorphism and body mass index (BMI) or six biochemical parameters, serum concentration of total cholesterol (TC), high density lipoprotein (HDL) cholesterol, triglyceride (TG), asparatate aminotransferase (AST), alanine aminotransferase (ALT), fasting plasma glucose. The genotype frequencies were observed at the frequencies of 24.3% for AA, 48.2% for AG and 27.5% for GG, respectively. When BMI and the biochemical parameters were compared by ANOVA among individuals with each genotype, the statistical difference was observed only for BMI (P=0.016). Bonferroni's test demonstrated that the men with the AG genotype have higher BMI than those with the AA genotype (22.4±2.8 vs. 21.4±2.2)(P=0.04). The individuals with the AG genotype also showed trend to have higher BMI than those with the GG, although the difference was not statistically apparent (22.4±2.8 vs. 21.5±2.3) (P=0.07). Our results indicated that the young healthy Japanese men with the AG heterozygote showed higher BMI than those with other genotypes. J. Med. Invest. 53 : 218-222, August, 2006

Keywords **:** *UCP-1, A-3826G polymorphism, Japanese, young male, obesity*

INTRODUCTION

Lifestyle-related disease including obesity, type 2 diabetes mellitus, and coronary artery disease is thought to be related to both genetic and environmental factors (1), and is becoming one of the most major health issues in the advanced nations all over the world including Japan (2, 3). There are many reports which point out that change in the dietary habits from Japanese to Western style is underlain in the background of the increase in lifestyle-related diseases in Japan (4).

The approach to the genetic background of lifestyle-related disease by recent molecular genetics revealed many genes related to the diseases (1). Those genes are classified to different categories such as growth factor, signal transduction, energy production and energy expenditure (1). So far, although many association studies on relationship between

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the genetic variations and lifestyle-related disease have been carried out, it is often reported that those results cannot be reproduced in the populations with different ethnicity or environmental factors (5, 6).

UCP-1 (Uncoupling protein-1), which is expressed in brown adipose tissue (BAT) important role for energy homeostasis, is a proton transporter that uncouples oxidative metabolism from ATP synthesis and energy consumption (7, 8). The UCP-1 gene is located on chromosome 4q31 and a single nucleotide polymorphism A-3826 G in the 5' flanking region has been already reported (9). The relationship between the A-3826G polymorphism and the physical or metabolic variables has been studied in many counties and many populations (10-17). There are reports that show that the individuals with the G allele of the A-3826G polymorphism have higher BMI than the individuals without the G allele (13, 17). Many of those observations are based on the comparison between the normal controls and the patients with lifestyle-related disease, and are often controversial (10-13, 15-17). Importantly, the frequency of the G allele is considerably different among different populations (13, 15, 16).

In this study, we show the genotype frequencies of the A-3826G polymorphism in the UCP-1 gene and describe relationship between the polymorphism and BMI or biochemical parameters in the young Japanese males.

SUBJECTS AND METHODS

Subjects

A total of 251 young Japanese males between the age of 22 and 29 (mean±SD, 22.7±1.6y) were recruited. They had no apparent medical problems. All participants were students of the University of Tokushima and gave informed consents.

This study was carried out as a part of our research on relationship between lifestyle-related disease and environmental or genetic factors, which had been approved by the ethical committee of The University of Tokushima.

Measurement of BMI and biochemical parameters

Blood samples were obtained from the subjects after an overnight fasting. Serum total cholesterol (TC), high density lipoprotein (HDL) cholesterol, triglyceride (TG), asparatate aminotransferase (AST), alanine aminotransferase (ALT) were automatically measured by Automatic analyzer 7150 (Hitachi High-Technologies Corp., Tokyo, Japan). Body mass index [weight (kg)/height (m^2)] was calculated from body weight and height.

Genotyping of the A-3826G polymorphism

Genomic DNA was extracted from peripheral blood leucocytes with QIAamp Kit (QIAGEN GmbH, Germany) according to the manufacture's instruction. The A-3826G polymorphism was detected according to the previous report (9). In brief, after polymerase chain reaction (PCR) was carried out to amplify the region containing the A-3826G polymorphism upstream the UCP-1 gene, the PCR products were digested with BclI endonuclease and were separated by electrophoresis with 2.5% agarose gel. The G allele gave one 470 bp fragment whereas the A allele gave 310 bp and 160 bp fragments.

Statistical analysis

Statistical analysis was performed with the SPSSver11 (SPSS Inc.). Differences between genotypes and means of BMI or biochemical parameters were analyzed with Bonferroni's test if ANOVA was significant. P value less than 0.05 were considered to be significant. Logarithmic transformation of ATL, AST and TG was performed to improve normality.

RESULTS AND DISCUSSION

The frequencies of three genotypes of A-3826G polymorphism in the UCP-1 gene were 61 (24.3%) for AA, 121 (48.2%) for AG and 69 (27.5%) for GG in 251 young healthy Japanese males, respectively. The genotype distribution is in agreement with the Hardy-Weinberg law (data is not shown). This distribution was similar to those previously reported in Japanese population (14) and in Korean population (15). The frequency of the G allele observed in this study was about 2- fold higher than in Finns study (16). Since the G allele is not common in many countries, researchers often compare biological parameters between individuals with the A allele and with the G allele rather than between individuals with each genotype (10, 11, 16). However, since the G allele is common in Japan as shown in the present and other studies, we could analyze relationship between three geno-

types and many parameters including BMI.

We compared the biological parameters including BMI and six biochemical parameters between the individuals with each genotype of the A-3826G polymorphism. The physical and metabolic characteristics of subjects according to the genotype of UCP-1 are summarized in Table 1. The significant differences by ANOVA were found only for BMI (P=0.016), while biochemical parameters showed no statistical differences. Although the biological parameters were also compared between the individuals with the A allele and with the G allele, the statistical difference was not obvious.

Since BMI among the young men with different genotypes were statistically different, we carried out Bonferroni's test and found that the individuals with the AG heterozygote have significantly higher BMI than those with the AA homozygote (P=0.04)(Fig.1). Moreover, BMI of the subjects with the AG heterozygote had trend to have higher BMI than with the GG homozygote, although no statistical difference was observed (P=0.07).

Table 1. Summary of characteristic of BMI and biochemical parameters in the individuals with each genotype of the A-3826G polymorphism of the UCP-1 gene.

Characteristics	Genotype			Total	P value#1
	A/A	A/G	G/G		
n#2	61(44)	121(101)	69(59)	251	
Age	22.8±1.6	22.6±1.6	22.7±1.5	22.7±1.6	0.77
BMI	21.37±2.23	22.4±2.75	21.49±2.29	21.91±2.55	0.016
Plasma glucose(mg/dl)	90.16±8.31	91.22±11.08	89.61±5.77	90.52±9.22	0.48
Total cholesterol(mg/dl)	183.16±29.41	178.16±26.71	177.75±29.37	179.59±28.09	0.56
HDL cholesterol(mg/dl)	62.52±11.75	58.58±11.75	60.17±11.65	59.89±11.68	0.17
Triglyceride(mg/dl)	79.48±49.68	82.6±50.86	73.9±38.47	79.45±47.43	0.46
ALT(U/L)	17.07±8.62	20.41±13.22	17.28±8.68	18.74±11.17	0.08
AST(U/L)	19.62±4.72	22.45±14.78	19.58±6.17	20.97±11.08	0.06

Data are expressed as the mean ±SD.
#1. P Values by ANOVA are shown.
#2. Numbers in the round brackets show those of samples analyzed for HDL choleterol.

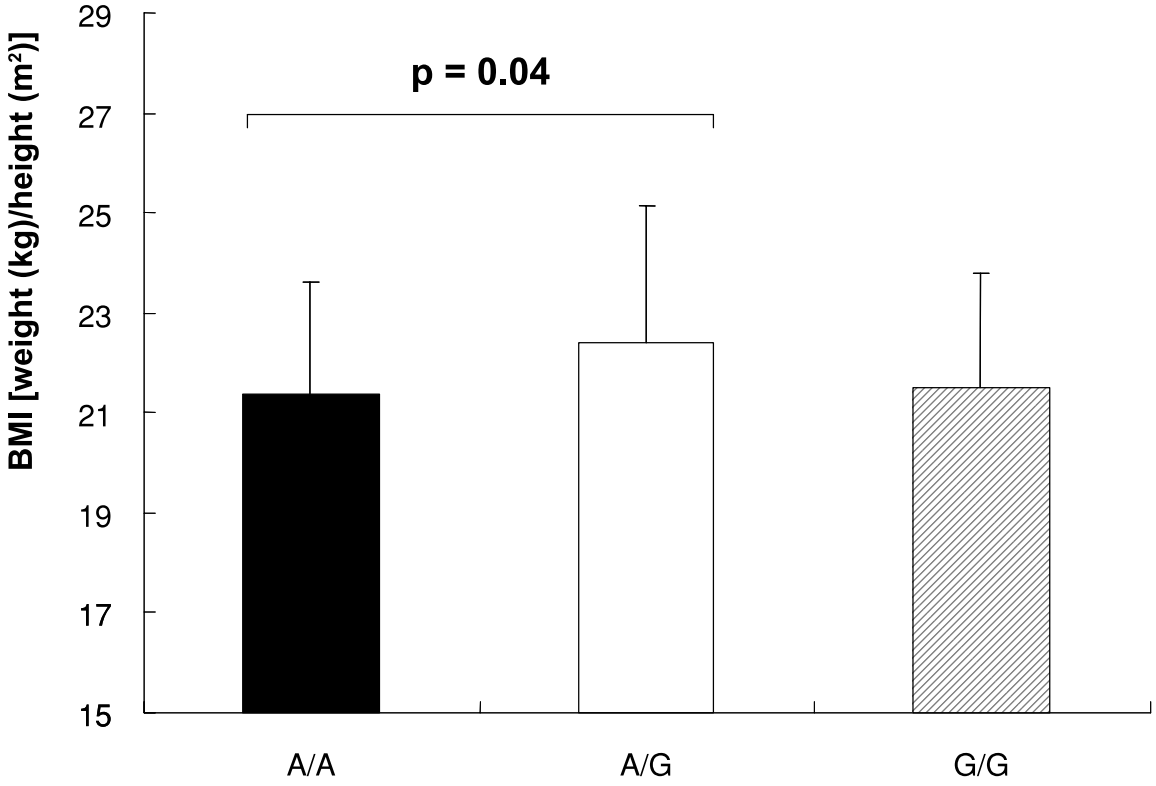

Figure 1: Comparison of the relationship between the individuals with each genotype of the A-3826G variation of the UCP-1 gene. The vertical scale shows value of BMI. Data are expressed as the mean±SD. P value is Bonferroni' test.

There is a report showing that the A-3826G polymorphism, which is located in the 5' flanking region of the UCP-1 gene, affects transcription of the UCP-1 gene (18). Therefore, it seems possible that the amount of the UCP-1 transcript is different between the young males with the AG heterozygote and those with the AA or with the GG homozygote. The expression level of the UCP-1 gene in the individuals with the AG genotype may be crucial for higher BMI.

To assess effect of the A-3826G polymorphism on BMI, differences in ethnicity and age are important. In this study, subjects were young Japanese between the age of 22 and 29. Ages of subjects in many other studies on the A-3826G polymorphism seem higher than those of the subjects in the present study. It is intriguing to investigate relationship between BMI and genotype frequencies of the A-3826G polymorphism in the UCP-1 gene in different populations with the ages similar to those of subjects in this study.

In conclusion, our results demonstrated that the combination of the A allele and the G allele of the A-3826G polymorphism in the UCP-1 gene is important for higher BMI in young Japanese males.

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