Peroxisome proliferator-activated receptor-gamma (PPARγ) is a factor in insulin resistance. In addition, PPARγ promotes the differentiation of small fat cells, which is accompanied by obesity. Recently, Spiegelman and colleagues (1) reported that PPARγ plays a role in obesity. In the obese state, pro-inflammatory signals lead to cleavage of the p35 protein to p25 (2), which activates CDK5 in the nucleus. Then, CDK5 phosphorylates PPARγ on serine 273, thereby preventing the transcription of specific PPARγ targets that have anti-obesity effects. Targeting PPARγ is now thought to be a logical strategy for combating obesity.

Single-nucleotide polymorphisms (SNPs) in the PPARγ gene also affect obesity. A recent genome-wide association study of 1,341 Framingham Heart Study participants revealed that six SNPs in the PPARγ gene or in its vicinity, rs2938392, rs709157, rs10510422, rs10510423, rs2454431, and rs963163, were associated with body mass index (BMI) and waist circumference (3). Another genome-wide study in a young population from Latin America, including Mexico, suggested that rs2938392 and rs1175542, or their haplotypes, influence BMI (4). These findings are important for the prevention and improvement of lipid metabolism disorders. We have previously reported that SNPs of fatty acid desaturase 1 are associated with plasma lipid profiles and could contribute to dyslipidemia in Japanese males. The aim of this study was to investigate the anti-obesity effects of PPARγ variants on lipid profiles. One hundred and thirty-eight (138) Japanese males participated in the study. Their serum lipid markers and the fatty acid composition of their red blood cell (RBC) membranes were determined. The stearoyl-CoA desaturase 1 (SCD1) indices were represented as the fatty acid product : precursor ratios. The participants were genotyped for the single-nucleotide polymorphism rs2938392 in the PPARγ gene. The participants’ fitness habits were also surveyed by questionnaire. The effects of habitual exercise on the measured lipid parameters were compared in each genotype group. No association between the genotypes in the PPARγ gene and the biochemical data was found. However, the serum triglyceride levels and the SCD1 indices in RBC membranes were significantly higher in the participants who carried the major rs2938392 allele (A/A) and did not habitually exercise than in those who did exercise. These findings indicate that the risk for detrimental lipid profiles in the absence of habitual exercise depends on the PPARγ genotype in Japanese males.

Key Words fatty acid, red blood cell, single nucleotide polymorphisms, stearoyl-CoA desaturase 1

Note

Efficacy of Habitual Exercise for Improving Lipid Profiles Depends on the PPARγ Genotype in Japanese Males

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Summary Peroxisome proliferator-activated receptor gamma (PPARγ) responds to thiazolidinedione derivatives, which are ligands of PPARγ, and affects insulin resistance. Recently, a PPARγ study reported that in high-fat-diet-induced obesity, the phosphorylation of PPARγ prevented the transcription of specific PPARγ targets that have anti-obesity effects. We previously reported that genetic variants of the fatty acid desaturase were associated with plasma lipid profiles and could contribute to dyslipidemia in Japanese males. The aim of this study was to investigate the anti-obesity effects of PPARγ variants on lipid profiles. One hundred and thirty-eight (138) Japanese males participated in the study. Their serum lipid markers and the fatty acid composition of their red blood cell (RBC) membranes were determined. The stearoyl-CoA desaturase 1 (SCD1) indices were represented as the fatty acid product : precursor ratios. The participants were genotyped for the single-nucleotide polymorphism rs2938392 in the PPARγ gene. The participants’ fitness habits were also surveyed by questionnaire. The effects of habitual exercise on the measured lipid parameters were compared in each genotype group. No association between the genotypes in the PPARγ gene and the biochemical data was found. However, the serum triglyceride levels and the SCD1 indices in RBC membranes were significantly higher in the participants who carried the major rs2938392 allele (A/A) and did not habitually exercise than in those who did exercise. These findings indicate that the risk for detrimental lipid profiles in the absence of habitual exercise depends on the PPARγ genotype in Japanese males.

Key Words fatty acid, red blood cell, single nucleotide polymorphisms, stearoyl-CoA desaturase 1

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acid (FA) desaturase, perilipin, and hormone-sensitive lipase genes alter obesity-inducible lipid profiles in Japanese males (5, 6). However, the association of obesity with PPARγ variants has not been clarified in Japanese males. Here, we focused on PPARγ as an anti-obesity target and investigated the PPARγ variant-specific effects on the participants’ lipid profiles.

Stearoyl-CoA desaturase 1 (SCD1; EC 1.14.99.5), a target gene of PPARγ, is known to regulate cholesterol and FA homeostasis. High SCD1 activity and alterations in the balance between monounsaturated and saturated FA have been implicated in various diseases, including heart disease, atherosclerosis, obesity, and insulin resistance (7–9). Therefore, SCD1 is thought to be an important therapeutic target for the development of anti-obesity drugs. In human studies, fatty acid product : precursor ratios in adipose tissue and various blood lipid fractions have been used to investigate the association between SCD1 and the risk of a number of diseases (10). The expression of SCD1 mRNA can be estimated from the fatty acid ratios in plasma and liver lipid fractions (11). Here, we also analyzed the association between estimated SCD1 levels and PPARγ variants.

Materials and Methods

Participants. Of the 150 participants initially recruited, the data of 12 participants were excluded for the following reasons: protocol violation (n = 2); missing data for weight, waist circumference, and exercise frequency (n = 4); and serum triglycerides (TG) ≥400 mg/dL (n = 6). Consequently, 138 participants (74 with waist circumferences <85 cm and 64 with waist circumferences ≥85 cm) were included in this study. Details regarding the characteristics of the participants were as previously described (5). The study protocol adhered to the Japanese Government’s Ethical Guidelines Regarding Epidemiological Studies, in accordance with the Declaration of Helsinki, and the study was approved by the Institutional Review Board of Human Genome Research Ethics at the Institute of Life and Environmental Science for Human Life, Ochanomizu University, Japan.

Measurement of clinical characteristics. All participants completed a questionnaire that included their fitness habits and medical histories. Answers concerning the fitness habits were ranked on a three-point Likert scale: 1, do not habitually exercise; 2, habitually perform light exercise; and 3, habitually perform hard exercise. Answer 1 was categorized as “No habitual exercise,” and 2 and 3 were categorized as “Habitual exercise.” The clinical characteristics of the participants were evaluated (5). After overnight fasting, blood samples were collected for multiple biochemical assays, measurement of FA composition and PPARγ genotyping. The multiple biochemical assays were performed as described in a previous report (5).

Genotyping of PPARγ gene polymorphisms. Based on the HapMap project data (http://www.hapmap.org), the SNP rs2938392 (location on the NCBI Assembly: 12434608, assay ID C_11908952_10) was genotyped using the TaqMan SNP allelic discrimination method with an ABI 7300 system (Applied Biosystems, Foster City, CA). Genomic DNA was extracted from 5 mL blood samples using a commercially available DNA extraction kit for blood samples (Takara, Kyoto, Japan) according to the manufacturer’s instructions.

Assessment of fatty acid composition. FA compositions of red blood cell (RBC) membranes were measured as described in a previous report (5). The intra-assay coefficients of variation of the analyzed FAs were C16:0, 0.66%; C16:1, 0.56%; C18:0, 0.54%; C18:1 (n-9), 0.46%; and C18:1 (n-7), 0.71%. The measured FA levels were expressed as percentages. The SCD1 desaturation indices represented the FA product : precursor ratios, as determined by the integrated area under the gas chromatogram peaks.

Statistical analysis. Biochemical data are shown as the mean±SD. Pearson’s chi-square test was used to compare the frequencies between participants with a normal waist circumference (<85 cm) and those with a large waist circumference (≥85 cm). The Hardy-Weinberg equilibrium test was used to evaluate the consistency between the observed and expected genotype frequencies. To examine the relationship among genotypes, a one-way ANOVA and a Bonferroni correction were conducted. To examine the relationship between habitual exercise and the biochemical data of the study participants, a Mann Whitney U-test was conducted. A two-sided p value of <0.05 was considered to be statistically significant. These statistical analyses were performed with SPSS (Statistical Package for the Social Sci-

Table 1. Genotype distribution frequencies.

<table>
<thead>
<tr>
<th>PPARγ A&gt;G rs2938392</th>
<th>Genotype (n=138)</th>
<th>Nucleotide frequency</th>
<th>p-value</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>AA</td>
<td>AG</td>
<td>GG</td>
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<tr>
<td>Frequency (number of persons)</td>
<td>46</td>
<td>65</td>
<td>27</td>
</tr>
<tr>
<td>Frequency rate (%)</td>
<td>32.4</td>
<td>49.1</td>
<td>18.6</td>
</tr>
<tr>
<td>Frequency rate calculated from observed values (%)</td>
<td>44.7</td>
<td>67.7</td>
<td>25.7</td>
</tr>
</tbody>
</table>

The average data for 138 participants are summarized.

Genotype frequencies were tested for Hardy-Weinberg equilibrium by the chi-squared test.

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Frequency (number of persons)</th>
<th>Frequency rate (%)</th>
<th>Frequency rate (%)</th>
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<tbody>
<tr>
<td></td>
<td>46</td>
<td>32.4</td>
<td>49.1</td>
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<td></td>
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</table>
Table 2. Relationships between the PPARγ rs2938392 genetic polymorphism (A/G) and the biochemical data.

<table>
<thead>
<tr>
<th>Clinical characteristics</th>
<th>AA</th>
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<th>B-value</th>
<th>AA</th>
<th></th>
<th></th>
<th></th>
<th>B-value</th>
<th>AA</th>
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<th></th>
<th></th>
<th>B-value</th>
<th>AA</th>
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<th>B-value</th>
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<th>B-value</th>
<th>AA</th>
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<th>B-value</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>Habitual exercise (n=20)</td>
<td>No habitual exercise (n=26)</td>
<td>Total (n=46)</td>
<td></td>
<td>Habitual exercise (n=31)</td>
<td>No habitual exercise (n=34)</td>
<td>Total (n=65)</td>
<td></td>
<td>Habitual exercise (n=13)</td>
<td>No habitual exercise (n=14)</td>
<td>Total (n=27)</td>
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<tr>
<td>Weight (kg)</td>
<td></td>
<td>69.8±7.3</td>
<td>67.6±10.0</td>
<td>68.5±8.9</td>
<td>0.272</td>
<td>67.8±7.8</td>
<td>69.8±9.4</td>
<td>68.8±8.7</td>
<td>0.510</td>
<td>67.1±10.1</td>
<td>69.4±7.6</td>
<td>68.3±8.8</td>
<td>0.325</td>
<td>0.961</td>
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<tr>
<td>BMI (kg/m²)</td>
<td></td>
<td>24.1±2.5</td>
<td>24.0±2.9</td>
<td>24.1±2.7</td>
<td>0.851</td>
<td>23.7±2.4</td>
<td>24.3±2.9</td>
<td>24.0±2.7</td>
<td>0.428</td>
<td>23.2±3.4</td>
<td>24.4±2.6</td>
<td>23.8±3.0</td>
<td>0.141</td>
<td>0.931</td>
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<tr>
<td>WC (cm)</td>
<td></td>
<td>83.9±6.0</td>
<td>85.4±6.5</td>
<td>84.7±6.3</td>
<td>0.417</td>
<td>82.7±5.3</td>
<td>85.1±6.8</td>
<td>83.9±6.2</td>
<td>0.156</td>
<td>82.4±4.5</td>
<td>83.3±4.8</td>
<td>82.9±4.6</td>
<td>0.647</td>
<td>0.461</td>
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<tr>
<td>Triglycerides (mg/dL)</td>
<td></td>
<td>144±102</td>
<td>192±89</td>
<td>171±97</td>
<td>0.041*</td>
<td>135±81</td>
<td>147±67</td>
<td>141±73</td>
<td>0.588</td>
<td>139±82</td>
<td>145±79</td>
<td>142±79</td>
<td>0.943</td>
<td>0.149</td>
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<tr>
<td>TC (mg/dL)</td>
<td></td>
<td>216±25</td>
<td>223±42</td>
<td>22±35</td>
<td>0.387</td>
<td>208±44</td>
<td>217±33</td>
<td>213±39</td>
<td>0.354</td>
<td>205±37</td>
<td>205±31</td>
<td>205±33</td>
<td>0.943</td>
<td>0.252</td>
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<tr>
<td>LDL-C (mg/dL)</td>
<td></td>
<td>127±21</td>
<td>129±42</td>
<td>128±34</td>
<td>0.782</td>
<td>130±38</td>
<td>136±29</td>
<td>133±34</td>
<td>0.337</td>
<td>124±38</td>
<td>121±28</td>
<td>122±33</td>
<td>0.905</td>
<td>0.356</td>
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Saturated and monounsaturated fatty acids

| C16:0                    |    | 29.3±6.3 | 25.4±5.0 | 27.1±5.9 | 0.088 | 25.5±4.3 | 24.9±6.8 | 25.2±5.7 | 0.486 | 31.3±15.1 | 28.0±6.5 | 29.6±11.4 | 0.519 | 0.027* |
| C16:1                    |    | 0.6±0.3  | 1.1±1.0  | 0.9±0.8  | 0.076 | 0.9±0.9  | 1.2±1.8  | 1.0±1.4  | 0.864 | 0.9±0.7  | 0.7±0.7  | 0.8±0.7  | 0.519 | 0.613 |
| C18:0                    |    | 20.2±3.5 | 18.3±6.1 | 19.1±5.2 | 0.025* | 18.1±2.9 | 18.0±7.0 | 18.0±5.4 | 0.684 | 18.0±5.5 | 19.1±5.4 | 18.6±5.3 | 0.943 | 0.565 |
| C18:1 (n=9)              |    | 15.8±3.2 | 19.1±6.3 | 17.7±5.4 | 0.088 | 16.1±3.6 | 17.4±6.9 | 16.8±5.6 | 0.495 | 16.0±4.7 | 16.2±5.2 | 16.1±4.9 | 0.867 | 0.465 |
| C18:1 (n=7)              |    | 1.3±0.3  | 1.6±0.8  | 1.5±0.6  | 0.063 | 1.4±0.4  | 1.7±1.0  | 1.6±0.8  | 0.401 | 1.5±0.6  | 1.5±0.7  | 1.5±0.6  | 0.336 | 0.809 |

SCD1 desaturase indices

| C16:1 (n=7)/C16:0        |    | 0.02±0.01 | 0.05±0.06 | 0.04±0.05 | 0.014* | 0.04±0.04 | 0.06±0.11 | 0.05±0.04 | 0.906 | 0.04±0.04 | 0.03±0.03 | 0.03±0.03 | 0.616 | 0.451 |
| C18:1 (n=9)/C18:0        |    | 0.8±0.3   | 1.3±1.2  | 1.1±0.9  | 0.026* | 0.9±0.4   | 1.4±1.7   | 1.2±1.3   | 0.984 | 1.0±0.5  | 1.0±0.6  | 1.0±0.6  | 0.905 | 0.702 |

Data are presented as the mean±standard deviation unless otherwise indicated. The averages of the clinical data of 138 persons are summarized. * Indicates significant differences at p<0.05 between “Habitual exercise” and “No habitual exercise” groups by the Mann-Whitney U-test. One-way ANOVA and Bonferroni correction were conducted among three genotypes. 

* Indicates a significant difference at p<0.05 by one-way ANOVA.

All participants completed a standard questionnaire that included exercise frequency. Answers were on a three-point Likert scale: 1, do not habitually exercise; 2, habitually perform light exercise; 3, habitually perform hard exercise. Answer 1 was categorized “No habitual exercise”, and 2 and 3 were categorized as “Habitual exercise.”
PPARγ Variants and the Exercise-Inducible Effect

Results and Discussion

Significant differences were not observed in the clinical characteristics or FA levels between the participants with a large waist circumference (≥85 cm) and those with a normal waist circumference (<85 cm), except for body weight, BMI and TG. Table 1 shows the genotype distributions and nucleotide frequencies in this study population. The minor allele frequency of the PPARγ SNP rs2938392 (A/G) was G=0.43. The allelic frequencies were in Hardy-Weinberg equilibrium. No significant difference in the frequencies of genotypes (p=0.504) was observed when the genotype distributions of participants with a waist circumference of <85 cm and those with a waist circumference of ≥85 cm were compared. Therefore, the waist circumference distribution was not related to the genotype distribution.

Table 2 shows the associations between PPARγ A>G rs2938392 genotypes and the biochemical data. Significant differences among the genotypes were not observed after Bonferroni correction of the biochemical data. Table 2 also shows the PPARγ variant-specific lipid profiles and biochemical data of the study participants who do not habitually exercise. The AA homozygotes who do not habitually exercise had significantly higher SCD1 indices (p=0.014, 0.026) and serum TG (p=0.041) than those who exercised habitually, whereas no differences were observed in the participants with the other genotypes (A/G or G/G). The AA homozygote participants who do not habitually exercise also tended to have higher monounsaturated FA levels [C16:1, C18:1 (n-9), C18:1 (n-7)] and lower saturated FA levels [C16:0, C18:0] than participants with the AA allele who habitually exercise. However, these effects were not found in the carriers of the other genotypes of rs2938392 (A/G or G/G).

Exercise-associated generation of PPARγ ligands activates PPARγ signaling events and upregulates genes related to lipid metabolism (12, 13). For example, in a previous study, sedentary participants in a low-intensity exercise program (walking 10,000 steps 3 times per week for 8 wk) exhibited increased monocyte PPARγ DNA-binding activity and enhanced expression of the PPARγ target genes CD36, PPARγ coactivator-1α (PGC-1α), and liver X receptor-α (LXRα) within monocytes and, consequently, exhibited upregulation of the LXRα target genes. ATP-binding cassette (ABC) subfamily A member 1 (ABCA1) and ABC subfamily G member 1 (ABCG1), known as the cholesterol efflux regulatory protein. These results might be related to different PPARγ activities resulting from the different genotypes of PPARγ SNPs. However, how PPARγ genotypes affect habitual exercise-induced improvement of lipid profiles remains unclear.

In contrast to a previous report that included individuals of European descent and the households of Framingham, Massachusetts, there was no association between BMI and rs2938392 in this study. The reason for this difference may be racial differences. For example, the frequencies of the immunoglobulin G haplotypes, which are markers of genetic distribution, vary widely between residents of the Japanese archipelago and Korean peninsula in East Asia (14). In spite of the northern Mongoloid pattern in both the Japanese and Korean groups, a highly significant heritable heterogeneity has been identified between the two populations (14). Therefore, it is worth examining the link between genetic variants and biomarkers in the Japanese population.

In this study, we used RBC membranes to investigate FA compositions. Some FAs in the RBC membrane are suitable biomarkers of long-term dietary intake (15). Additionally, trans-fatty acids in the RBC membrane reflect dietary trans-fat intake, as measured by a food frequency questionnaire, more strongly than serum trans-fatty acids (16).

Several limitations of this study should be addressed. First, this study was restricted to men of only one racial group, and the sample size may not have captured a representative study population. Second, the fitness habits of the participants were measured by questionnaire, which made it difficult to assess the precise intensity of exercise. This may have limited our ability to detect associations between the intensity of exercise and the FA compositions of RBC membranes. However, the results of the present study are novel and provide further evidence that habitual exercise improves the balance among several FA levels and aids in recuperation from diseases caused by metabolic disorders characterized by unbalanced FAs.

In conclusion, our results provide evidence that a lack of habitual exercise increases SCD1 desaturation and normal TG levels, depending on the genetic variant of PPARγ. These results indicate that the risk for detrimental lipid profiles in those who perform no habitual exercise depends on the PPARγ genotype. Further studies are recommended to investigate the associations between habitual exercise and the serum lipid profile/FA composition.

Acknowledgments

We gratefully acknowledge the staff of the Fukuoka Institute of Occupational Health for data collection and for their excellent and dedicated work.

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Conflicts of Interest

None to declare.

Disclosure

The authors declare that they have no competing interests regarding the manuscript.
REFERENCES


